CITRUS RESPONSE FUNCTIONS TO N, P, AND K FERTILIZATION AND N UPTAKE DYNAMICS

By

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To my wife Patricia and my daughter Ana Clara who have embraced me with a tender feeling, I dedicate all my achievements.

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The citrus industries in Brazil and Florida account for up to 49% of the total world production. To maintain a competitive edge in the world market, and to respond to the public pressure to minimize the adverse environmental impact of non-point source pollution of nutrients into groundwater, it has been increasingly important to develop nutrient management recommendations to improve nutrient uptake efficiency, minimize nutrient losses and reduce the impact on ground water.

Since current recommendations have paid little attention to nutrient fate studies, responses of nonbearing trees (<5-yr-old) to NPK fertilization in Brazil were investigated. Furthermore, the increasing use of different rootstocks has justified the need for determining the nutritional requirements of trees on new rootstocks.

Field studies showed that rootstocks affected the response of nonbearing orange trees to NPK fertilization. Fertilizer rates for maximum tree growth and fruit yield were

greater than those currently used in Brazil. The critical nutrient concentrations in the soil and leaves may need to be reevaluated. Data show that the critical levels of P in the soil, and that of N in the leaf tissue, are greater than those currently recommended for bearing trees. Orange trees on Rangpur lime were less responsive to fertilization than those on Cleopatra mandarin (Cleo) or Swingle citrumelo rootstocks. Fruit yield of trees on Cleo increased with P rates (up to 2200 g P_2O_5 per tree), while yield of trees on Swingle increased significantly with K rates (up to 1800 g K_2O per tree) during a 5-yr fertilization program.

The results of studies conducted in a sandy Entisol (pH = 7.0) under 6-yr-old citrus trees in Florida demonstrated that ammonia volatilization accounted for 33% of applied urea. Similar results were observed in Brazil, even though soil pH was about 5.4. Nitrogen fertilization affected N mineralization and microbial biomass N as measured in 30-day period after fertilization. Leaching of N below the soil surface was low using a monitored under-the-tree low volume irrigation system. Recovery of ^{15}N by citrus trees from labeled ^{15}N fertilizers applied in the spring to the surface of the soil was 25.5% for urea and 39.5% for ammonium nitrate, 280 days after fertilization.

CHAPTER 1
INTRODUCTION

The Citrus Industry in Brazil and Florida

The term citrus is used to designate plants of the genus *Citrus*, and other related genera or hybrids that represent trees and shrubs of the Rutaceae family that bear oranges, mandarins, lemons, limes, grapefruits or other such fruit. Orange production represents about 70% of all citrus throughout the world (Food and Agriculture Organization, 1999).

Brazil is the leading producer of oranges in the world (22.7 million tons in 1999) followed by Florida (7.6 million tons in 1999). The combined production accounts for up to 49% of the overall world production (FAO, 1999; Florida Department of Citrus, 2000). The great majority of the Brazilian citrus growing area is in São Paulo State, which is located between 20 and 25°S longitude. In the São Paulo State plateau the average temperature varies between 20 and 23°C with mean annual rainfall of up to 1350 mm. Drought periods are common during the winter. The lower third of the state is below the Tropic of Capricorn, with mild weather of average temperature below 20°C and no water deficit.

About 75 to 80% of the São Paulo orange production is processed as frozen concentrated orange juice (FCOJ) at 65°Brix and exported to the USA, Europe and Asia. São Paulo exported 1.24 million metric tons of FCOJ in the 1998/99 season at an average

FOB price of US$ 1,430.00 ton^{-1}. The Florida orange production is 93% processed as FCOJ or chilled juice.

Citrus production makes a valuable contribution to the economy of Brazil and Florida considering the number of orange trees and area under production in São Paulo (210.7 million and 870,000 ha, respectively) and Florida (85.4 million and 266,700 ha, respectively) (Fundecitrus, 1998; Florida, 2000). The above statistics suggest a large demand for technology and agrochemicals (i.e., the cost of fertilizers and pesticides account for up to US$400 to 500 million annually in São Paulo), machinery, labor, and market costs. The commercialization of valuable by-products such as citrus peel oil and essences for the chemical industry (food flavoring, fragrances and solvents), and dried pulp pellets for animal feed constitute other sources of revenue from the citrus industry.

Citrus in São Paulo usually is produced in nonirrigated areas, on a range of soils, which include sandy (<10% clay) to clay-type soils (50 to 75% clay) such as Oxisols, Entisols, Alfisols, and Ultisols. Most of these soils are inherently low in fertility, very acid, very deep, and well drained.

In Florida, citrus is irrigated and grown in well-drained sands low in mineral nutrients and organic matter (Entisols), in fine-textured lowland soils with a sandy surface, often developed on calcareous marine deposits, with relative greater retention of nutrients (Alfisols), or in sandy, acidic, coarse-textured and poorly drained soils (Spodosols) (Alva and Tucker, 1999).

Soil physical and chemical conditions have a major impact on citrus production and thus, require continued research to develop soil management prescription to eliminate limiting factors and to optimize the production and quality.

Soil Fertility and Citrus Nutrition

Need for Research in São Paulo

Recent NPK recommendations for the São Paulo State citrus industry (Grupo Paulista, 1994) are based on criteria developed by Cantarella et al. (1992) with field trials carried out during 1986-1991. Prior to this long-term research, very little work had been conducted to evaluate citrus tree response to NPK fertilization (Rodriguez et al., 1965; Rodriguez and Moreira, 1969; Cruz et al., 1971; Malavolta et al., 1996). The previous recommendations of Grupo Paulista (1990) were reviewed, and the new recommendation included an increase in N and P rates, and a decrease in K. Soil nutrient levels were calibrated to citrus production response. Much of the attention was on the fertilization need for bearing trees, with very little information for nonbearing trees.

A recent estimate shows a total of 40 million nonbearing trees (<5-yr-old) in São Paulo State (Fundecitrus, 1998). At present, Rangpur lime (*Citrus limonia* Osb.) is the most commonly used rootstock. There has been an increase in the use of other rootstocks, such as Cleopatra (*C. reshni* hort. ex Tanaka) and Sunki (*C. sunki* hort. ex Tanaka) mandarins because of their tolerance to citrus blight (Timmer, 1988), and Swingle citrumelo (*Poncirus trifoliata* (L.) Raf. x *C. paradisi* Macf.) because of its resistance to *Phytophthora* bark infection and tolerance to root rot (Castle et al., 1993). In addition, trees on Swingle citrumelo have produced increased fruit yields with favorable attributes such as greater fruit size, juice color, and quality (Castle et al., 1988). While the use of different rootstocks has increased over the recent years, little is known about the nutritional requirements of the trees on different rootstocks.

Need for Research in Florida

Water quality concerns have come to the forefront with respect to Florida citrus industry, because of an increased number of wells, mostly shallow drinking water wells, with nitrate concentration in excess of the U.S. Environmental Protection Agency's recommended maximum contaminant limit (10 mg NO_3-N L^{-1}) for drinking water standards in some parts of citrus production regions. Embleton and Jones (1977) and Embleton et al. (1978) discussed the same issue in California during the 1980's. However, only recently have these issues attracted public attention in Florida.

Since the past fertilizer recommendations were primarily based on the horticultural response of citrus trees, there is a need to reevaluate the nutrient management recommendations in an effort to optimize the nutrient uptake efficiency and minimize losses, to address the horticultural response, water quality control, and economics of production.

To develop modified nutrient management recommendations, new databases have been generated from the following studies: (i) N diagnostic tools (Obreza, 1990), (ii) N management and fertilization that included issues on N leaching losses (Willis et al., 1990), fertigation (Willis et al., 1991), fertilizer reactions in the soil (Khakural and Alva, 1995), wastewater use (Maurer et al., 1995), foliar fertilization (Lea-Cox and Syvertsen, 1995), leaf N analysis and leaching (Alva and Paramasivam, 1998), and tree root distribution (Zhang et al., 1996, 1998), (iii) N uptake and distribution (Lea-Cox and Syvertsen, 1996; Syvertsen and Smith, 1996), (iv) N sources (Dou and Alva, 1998b), (v) N mineralization (Dou et al., 1997; Dou and Alva, 1998a), (vi) N dynamics in the soil system such as adsorption, transport (Li et al., 1995; McNeal et al., 1995), and

denitrification (Paramasivam et al., 1999), and (vii) cultural practices such as irrigation (Alva and Fares, 1998; Fares and Alva, 1999).

Research Goals

Modern agriculture is facing considerable environmental challenges to minimize the impact of agricultural products on soils, water, and air quality. Nutrient management programs have to be developed to supply crops with adequate quantities of plant nutrients to support maximum productivity and profitability while minimizing the adverse environmental impact.

The major objective of this study was to develop basis for nutrient management recommendations for nonbearing citrus trees and to investigate the nitrogen dynamics in the soil-plant system. Research was conducted in different ecological regions in São Paulo State, and in Polk County, Florida. The soil used in the Florida study was representative of light textured, sandy (>95% sand) Entisols with no containing layers thus vulnerable to excessive leaching of water and agrochemicals.

The overall objectives of the proposed study in São Paulo State, Brazil, were primarily to investigate recommendations for the NPK fertilization of nonbearing trees (<5-yr-old). Differences in nutritional requirements of the most common tree rootstocks used in the local citrus industry were studied. The dynamics of N fertilizer sources applied to the soil-plant system, including N usage and distribution in orange trees in Florida was investigated to increase nutrient uptake efficiency and minimize losses.

The specific objectives were to (i) develop basis for optimal nitrogen, phosphorus and potassium rates for nonbearing trees growth and production; (ii) investigate the

influence of citrus rootstocks on tree response to fertilization; (iii) develop guidelines for the use of soil and leaf analysis as bases for fertilizer recommendations; (iv) evaluate the gaseous loss of N by ammonia volatilization; (v) evaluate soil net N-mineralization; (vi) investigate partitioning of soil applied-^{15}N during early spring in different plant components at the fruit harvest.

CHAPTER 2
RESPONSE FUNCTIONS FOR YOUNG CITRUS TREES ON DIFFERENT ROOTSTOCKS TO N, P, AND K FERTILIZATION

Introduction

The majority of the Brazilian citrus growing area is located in the state of São Paulo with 210 million trees, of which about 40 million trees are less than 5-yr-old (Fundecitrus, 1998). Despite the significance of the local citrus industry to the economy of the state and the country, very little work has been conducted to evaluate the citrus tree response to NPK fertilization, especially for nonbearing trees in the field (Rodriguez et al., 1965; Rodriguez and Moreira, 1969; Cruz et al., 1971; Rodriguez et al., 1973).

Current guidelines on the NPK fertilization for the citrus trees in Brazil (Grupo Paulista, 1994) are defined for nonbearing (<5-yr-old) and producing trees and mostly based on soil and leaf analysis, and fruit yield criteria. Although recommendations are well established for producing trees, the same is not true for nonbearing trees. Nutritional requirements of nonbearing trees are expected to be different from those of producing trees because of greater growth rate and lower fruit yields. Rates of nitrogen are recommended based on tree age and range from 80 to 400 g N per tree per year, while rates for phosphorus or potassium are recommended based on soil analysis and range from 0 to 300 g P_2O_5 and 0 to 400 g K_2O per tree per year, respectively.

At present, Rangpur lime (*Citrus limonia* Osb.) is the most commonly used rootstock in the Brazilian citriculture. However, since this rootstock is sensitive to citrus

decline, a disease closely related to blight in Florida (Wutscher et al., 1980), there has been an increase in the use of others such as Cleopatra (*C. reshni hort.* ex Tanaka) and Sunki (*C. sunki* hort. ex Tanaka) mandarins because of their tolerance to this disease (Timmer, 1988). Swingle citrumelo (*Poncirus trifoliata* (L.) Raf. x *C. paradisi* Macf.) is considered highly resistant to *Phytophthora* bark infection and tolerant to root rot (Castle et al., 1993). In addition, trees on this rootstock have produced fruit with favorable attributes such as size, and juice color and quality (Castle et al., 1988).

While the use of different rootstocks has increased over the recent years, little is known about the nutritional requirements of the citrus trees on different rootstocks. Observations on commercial groves have shown differences in growth habit of the trees depending on the rootstock. New groves with trees grafted on Cleopatra mandarin that received high nitrogen fertilization showed greater vegetative growth without appreciable increase in fruit yields. Another problem observed is related to excess K that tends to promote poor vegetative growth.

Recommendations made in Florida for nonbearing citrus trees (<3-yr-old) are based on studies conducted at several locations (Rasmussen and Smith, 1961; Anderson and Calvert, 1967; Anderson and Martin, 1969; Calvert, 1969; Jackson and Davies, 1984; Ferguson et al., 1988, 1990; Obreza, 1990; Obreza and Rouse, 1991, 1993; Obreza, 1994; Willis et al., 1990; Willis and Davies, 1991) and include a range of rates for each tree age. Nitrogen rates recommended vary from 70 to 400 g N per tree per year, whereas phosphorus and potassium rates generally follow the same rate as for N for trees planted on previously uncropped soils; in previously cropped soils the rates of phosphorus may be reduced or omitted if soil test results indicate adequate P levels (Ferguson et al., 1995). Even though fertilizer guidelines make reference to a number of factors that influence

fertilizer requirements (i.e., N level of nursery trees, soil type, land history, fertilizer placement, application frequency and timing), the current fertilizer recommendations do not take into account the differences in citrus scion and rootstock.

The objectives of this study were to (i) establish nitrogen, phosphorus and potassium rates for maximum growth and fruit yield of nonbearing trees (<5-yr-old) in the State of São Paulo, Brazil; (ii) evaluate the differential response of scion/rootstock combinations to NPK fertilization, and (iii) evaluate the use of soil and leaf analysis guidelines as a basis for fertilizer recommendations.

Material and Methods

Characteristics of Study Areas

Three citrus groves with (i) 'Pera' sweet orange (*Citrus sinensis* (L.) Osb.) on Rangpur lime rootstock (*C. limonia* Osb.), (ii) 'Valencia' sweet orange (*C. sinensis* (L) Osb.) on Rangpur lime and Cleopatra mandarin (*C. reshni* hort. ex Tanaka) rootstocks, and (iii) 'Natal' sweet orange (*C. sinensis* (L) Osb.) on Rangpur lime, Cleopatra mandarin and Swingle citrumelo (*Poncirus trifoliata* (L.) Raf. x *C. paradisi* Macf.) rootstocks were used in this study. The experiments were conducted in the main citrus producing areas of the State of São Paulo, Brazil. The experimental sites are described in Table 2.1. Santa Cruz do Rio Pardo is located in the southeast region of the state and has an average temperature below 20°C and no water deficit with mean annual rainfall of 1760 mm. The soil was an Alfisol previously cultivated with eucalyptus. The other sites were located in the central and northwest regions with average temperature between 20 and 23°C and mean annual rainfall of 1350 mm. Drought periods are common during the

winter in these regions. In Bebedouro, the soil was an Alfisol and in Matão, an Oxisol previously cultivated with citrus and corn, respectively. Selected chemical properties of soils prior to the tree planting are presented in Table 2.2. Soils at each site received dolomitic lime application in amounts calculated to raise base saturation (0 to 20 cm depth layer) to approximately 70% (Quaggio et al., 1992).

Experimental Design

Experiments were arranged in a fractional factorial design of the ½ (4^3) type, with a total of 32 treatments, as proposed by Colwell (1978), and selected by Andrade and Noleto (1986). A residual mean square was obtained through the high-order interactions that provided an upper limit for the value of the error, and since such interactions were small in comparison with the error, they were used to provide an estimate of the experimental error (John and Quenouille, 1977). The experimental design consisted of only a selected fraction of the factorial combinations (Table 2.3). Confounding of unlike treatment effects was used to reduce the size of the experiment, increasing the efficiency of the model to measure meaningful effects (Colwell, 1994).

Treatments consisted of four N, P, or K rates calculated to be applied during five years after tree planting: N (400, 1000, 1600 and 2200 g N per tree, as ammonium nitrate, 33% N), P (400, 1000, 1600 and 2200 g P_2O_5 per tree, as triple superphosphate, 41% P_2O_5), and K (300, 800, 1300 and 1800 g K_2O per tree, as potassium chloride, 60% K_2O) (Table 2.3). The annual rates of N, P, and K were adjusted to account for the tree age as shown in Table 2.4. Fertilizer mixtures were manually applied on the soil surface and around the tree about 50 to 200 cm away from the trunk. Total annual rate of P was applied in a single dose in early spring, while those for N and K rates were divided into

Table 2.1. Some background information on the field experiments.

Location	Planting date	Scion	Rootstock †	Area	Spacing
				ha	m
Sta. Cruz R. Pardo	Jun. 1995	'Pera'	RL	1.2	7.0 x 3.0
Bebedouro	Mar. 1995	'Valencia'	RL, CL	1.4	7.0 x 3.5
Matão	Nov. 1994	'Natal'	RL, CL, SW	2.0	7.0 x 4.0

† RL = Rangpur lime, CL = Cleopatra mandarin, and SW = Swingle citrumelo rootstocks.

three applications from spring to fall. Micronutrients, including Zn, Mn, and B, were applied by foliar spray as recommended by Grupo Paulista (1994).

The experimental plots consisted of 5 uniform trees with the middle three used for sampling. Soil samples were taken from 0 to 20 cm depth layer in the 5th year after tree planting for determination of pH (0.01 mol L^{-1} $CaCl_2$), resin extractable P and K, and base saturation (V%) using the methods described by Raij et al. (1987). Leaf samples were collected from fruiting terminals in the 4th and 5th years for analysis of various mineral elements according to Bataglia et al. (1983). Tree growth measurements were conducted and canopy volume was calculated using the following expression: $V = 2/3\ \pi\ r^2\ h$, where V = canopy volume, r = canopy radius, and h = canopy height. Fruit yield was computed annually by summing up the weight of fruit, if more than one harvest per year were necessary. Fruits were evaluated for juice quality, including total soluble solids, acidity, and total soluble solids/acidity ratio, following methods described by Reed et al. (1986). Fruit harvest occurred at maturity, when total soluble solids/acidity was in the range of 12 to 14, and acidity < 0.8% (w/v).

Table 2.2. Selected chemical characteristics of soils at the different experimental sites before tree planting.

Local	Depth	P-resin	OM	pH †	K	Ca	Mg	H+Al	BS ‡
	cm	mg dm^{-3}	g kg^{-1}		----------	$mmol_c$ dm^{-3}	----------		%
Sta. Cruz R. Pardo									
	0-20	22	23	5.9	0.9	58	39	20	83
	20-40	6	11	4.2	0.4	9	7	41	29
Bebedouro									
	0-20	9	16	5.5	1.4	13	12	20	63
	20-40	5	11	4.6	1.1	14	6	30	34
Matão									
	0-20	10	24	5.3	2.0	17	10	25	53
	20-40	3	18	4.7	0.6	8	6	36	28

† Soil / $CaCl_2$ 0.01 mol L^{-1}, 1:2.5 ratio (v/v).
‡ Soil base saturation.

Data Analysis

Data were tested for significant differences among treatments using a randomized complete block ANOVA. Response functions of the type:

$Y = b_0+b_1N+b_2N^2+b_3P+b_4P^2+b_5K+b_6K^2+b_7NP+b_8NK+b_9PK$ were computed for each experiment, where Y is the dependent variable, b_0 to b_9 are the regression coefficients, and N, P and K are the total rates of N, P_2O_5 and K_2O applied during 5 years, by using the GLM procedure of the SAS® system (1996). Dependent variables were calculated as (i) the average of canopy diameter, height and volume, and leaf nutrient concentrations for the 4^{th} and 5^{th} years after tree planting, and (ii) the cumulative fruit yield for the same period.

Table 2.3. Treatment description and total nutrient rates applied during five years after tree planting.

Treatments	Nutrients		
	N	P_2O_5	K_2O
Block I		g per tree	
111	400	400	300
122	400	1000	800
133	400	1600	1300
144	400	2200	1800
212	1000	400	800
221	1000	1000	300
234	1000	1600	1800
243	1000	2200	1300
313	1600	400	1300
324	1600	1000	1800
331	1600	1600	300
342	1600	2200	800
414	2200	400	1800
423	2200	1000	1300
432	2200	1600	800
441	2200	2200	300
Block II			
114	400	400	1800
123	400	1000	1300
132	400	1600	800
141	400	2200	300
213	1000	400	1300
224	1000	1000	1300
231	1000	1600	300
242	1000	2200	800
312	1600	400	800
321	1600	1000	300
334	1600	1600	1300
343	1600	2200	1300
411	2200	400	300
422	2200	1000	800
433	2200	1600	1300
444	2200	2200	1300

Table 2.4. Nitrogen, phosphorus and potassium rates applied on an annual basis, during five years of citrus tree growth.

Total rate	Year after tree planting				
	1	2	3	4	5
g per tree			g N plant^{-1}		
400	28	56	72	104	140
1000	70	140	180	260	350
1600	112	224	288	416	560
2200	154	308	396	572	770
			g P_2O_5 plant^{-1}		
400	28	56	72	104	140
1000	70	140	180	260	350
1600	112	224	288	416	560
2200	154	308	396	572	770
			g K_2O plant^{-1}		
300	21	42	54	78	105
800	56	112	144	208	280
1300	91	182	234	338	455
1800	126	252	324	468	630

Results and Discussion

Soil Chemical Characteristics

The addition of nitrogen fertilizer acidified the soil surface layer (0-20 cm) after 5 years of treatment. Average decrease on soil pH was about 0.5 unit (Table 2.5). Such effect is due to an input of hydronium ions in the soil solution as a result of nitrification, and also to the leaching of basic cations to deeper soil layers. These combined processes caused a decrease on soil base saturation (V%) in the 0-20 cm layer in all experimental sites. Levels of exchangeable K were lower at higher rates of the nitrogen fertilization (i.e., N = 2200 g per tree) in Sta. Cruz R. Pardo and Matão, as estimated by the surface response models (data not shown). He et al. (1998) found that N fertilizer applied in a Florida Entisol with 26-yr-old white Marsh grapefruit resulted in a decrease of the soil pH (water/soil ratio, 2:1 v/w) by 0.7 to 1.7 unit after four years of fertilization. Their study also found an increase in leaching of P and K below the grapefruit trees rootzone with a decrease in soil pH. Panzenhagen et al. (1999) also reported a decrease of soil pH (about 1.1 unit) after 8 years of continuous application of N fertilizer to an Ultisol in Brazil with 'Montenegrina' tangerines.

The phosphorus fertilization increased plant available P in the soil to levels above 20 mg dm^{-3} (Table 2.5). Extractable soil P and K increased in a near-linear manner at 0-20 cm depth as the fertilizer rate increased in a short term experiment conducted with young 'Hamlin' orange trees in southwest Florida (Obreza, 1990). Similar effects were reported by Sobral et al. (2000) in an Oxisol under 'Pera' sweet orange in Brazil after 3 and 11 years of P and K fertilization. Lower increments on soil P were observed in

Table 2.5. Effects of fertilizer rates on selected soil chemical characteristics in the fifth year of NPK fertilization.

Dependent variable †	Fertilizer rate ‡ 1	2	3	4	Condition ‡§	F test	CV %
			Sta. Cruz R. Pardo				
pH	5.1	4.7	4.7	5.0	P = 1; K = 1	N_L*	6.5
V, %	50	34	32	43	P = 1; K = 1	N_L**	22.2
P-res, mg dm^{-3}	63	83	104	125	N = 1; K = 1	P_L**	62.0
K, $mmol_c$ dm^{-3}	0.8	1.3	1.7	2.0	N = 1; P = 1	K_L**	30.3
			Bebedouro				
pH	4.7	4.4	4.3	4.2	P = 1; K = 1	N_L**; N_Q*; NP*; PK**	4.1
V, %	36	28	24	24	P = 1; K = 1	N_L**; N_Q*; NP*; PK**	15.9
P-res, mg dm^{-3}	22	47	61	64	N = 1; K = 1	P_L**	58.7
K, $mmol_c$ dm^{-3}	1.7	2.7	3.5	4.2	N = 1; P = 1	K_L**	37.7
			Matão				
pH	4.8	4.6	4.5	4.5	P = 1; K = 1	N_L**; N_Q*	3.8
V, %	47	41	38	42	P = 1; K = 1	N_L**; N_Q*	15.4
P-res, mg dm^{-3}	44	62	72	75	N = 1; K = 1	P_L**	45.3
K, $mmol_c$ dm^{-3}	1.4	2.7	3.9	5.1	P = 1; N = 1	N_L**; K_L**; NK*	24.0

† P-res = anionic exchangeable resin; K = exchangeable potassium; pH = soil / $CaCl_2$ 0.01 mol L^{-1}, 1:2.5 ratio (v/v); V= base saturation.

‡ Rates 1, 2, 3, and 4 for N or P are 400, 1000, 1600 and 2200 g of N or g P_2O_5 per tree, respectively. Rates 1, 2, 3, and 4 for K are 300, 800, 1300, or 1800 g K_2O per tree.

§ Condition used to simplify response models. Subscript L or Q = linear or quadratic component of the response model, respectively.

*, ** Significant at P = 0.05 and 0.01, respectively.

Bebedouro, where specific adsorption of P was considered to be more important, since predominant minerals in this soil are aluminum oxides. Phosphorus fertilization also affected soil base saturation in Bebedouro (i.e., response model showed significant positive NP and PK terms). The specific adsorption of P into aluminum oxides is reported to increase negative charges of the exchange complex creating additional sites for exchange of basic cations (Uehara and Gillman, 1981).

Potassium fertilization significantly increased soil exchangeable K levels in all experimental sites (Table 2.5). Obreza (1990) verified that a small K supply accumulated in soils under citrus trees even though K is considered a relatively mobile nutrient in sandy soils of southwest Florida. Rates greater than 300 g K_2O per tree increased exchangeable K above initial levels observed in the 0-20 cm soil depth before tree planting (Table 2.5). A wider range of soil exchangeable K levels was found in the soil in Sta. Cruz R. Pardo location, where the soil texture is coarser and the rainfall is abundant throughout the year. In this situation, leaching of K is significant. The interaction of NK was evident in Matão, where at the 400 g N per tree, maximum exchangeable K in the soil was 5.1 $mmol_c\ dm^{-3}$, whereas at the 2200 g N per tree, maximum level for K was 3.6 $mmol_c\ dm^{-3}$, as estimated by the response model (data not shown).

Tree Growth

The coefficient of correlation determined for pooled averages ($n = 32$) of canopy volume and diameter across rootstocks was 0.93 ($P<0.0001$), whereas that for canopy volume and height was 0.82 ($P<0.0001$). Tree growth increased from the third to the fifth year after tree planting in the field with average canopy volume for all sites of 4.2 and 19.9 m^3, respectively (Table 2.6).

The experiments in Bebedouro and Sta. Cruz R. Pardo showed that rootstocks affected tree growth (Table 2.6). The trees on Cleopatra mandarin rootstock had greater vigor, and larger canopy (17.9 m^3; 5-yr-old trees) as compared to those on Rangpur lime (12.8 m^3; 5-yr-old trees). On the other hand, Swingle citrumelo rootstock has a semi-dwarfing effect on sweet orange trees (Davies and Albrigo, 1994). In Matão, trees on Rangpur lime and Cleopatra mandarin rootstocks produced vigorous canopy as compared to those on same rootstocks in Bebedouro. More frequent water deficit was probably restrictive to greater growth of trees on the Cleopatra mandarin in Matão. Trees on Cleopatra are intermediate drought tolerant (Davies and Albrigo, 1994), as well as trees on Swingle citrumelo (Hutchinson, 1974).

The NPK fertilization influenced tree growth, especially the average canopy volume in Sta. Cruz R. Pardo, where significant linear and quadratic responses ($P<0.01$) to the K and N fertilization were observed (Table 2.7). Model coefficients (Table 2.8) show a large positive contribution of K rates on canopy volume (i.e., 16.6 and 20.2 m^3 at 300 or 1800 g K_2O per tree, respectively; with P_2O_5 = 400 and N = 2200 g per tree). Soil acidification caused by N fertilization, as showed by decreased soil pH (Table 2.5) was inversely correlated with canopy growth (data not shown). Site characteristics in Sta. Cruz R. Pardo, such as low soil exchangeable K, high base saturation before tree planting and rainfall distribution, were major factors that defined the magnitude of such effects.

Tree growth response was less clear in Bebedouro (Table 2.7) since only average canopy diameter was significantly affected by treatments either for trees on Rangpur lime or Cleopatra mandarin rootstocks. The coefficients for NP (Rangpur lime) or N and P (Cleopatra mandarin) of the response models are small (Table 2.9). Other factors, such

Table 2.6. Overall mean canopy volume of citrus trees in different sites.

Tree †	Years after tree planting		
	3	4	5
	---------- m^3 ----------		
		Sta. Cruz R. Pardo	
P/RL	5.6 (0.7‡)	11.7 (1.7)	23.1 (2.0)
		Bebedouro	
V/RL	3.3 (0.5)	7.9 (1.3)	12.8 (2.0)
V/CL	3.2 (0.2)	8.8 (1.6)	17.9 (2.7)
		Matão	
N/RL	6.3 (1.0)	9.8 (2.7)	28.3 (3.7)
N/CL	4.4 (0.9)	8.6 (2.6)	24.3 (4.7)
N/SW	2.5 (0.7)	5.5 (2.4)	13.5 (2.8)
Mean	4.2	8.7	19.9

† P = 'Pera', V = 'Valencia', and N = 'Natal' sweet oranges; RL = Rangpur lime, CL = Cleopatra mandarin, and SW = Swingle citrumelo rootstocks.

‡ The standard error of the mean (n = 32).

Table 2.7. Summary of the analysis of variance for dependent variables evaluated on the 4th and 5th years after tree planting at different experimental sites.

Tree †	Dependent variable	Mean ‡	F test §	CV, %
		Sta. Cruz R. Pardo		
P/RL	canopy diameter, cm	328	N_Q**	3.2
	canopy height, cm	297	N_Q*, K_L**	3.1
	canopy volume, m^3	17.4	N_Q**, K_L**	8.0
	fruit yield, t ha^{-1}	69.0	K_L**, NP**, PK*	8.4
	leaf N, g kg^{-1}	29.8	NS	4.8
	leaf K, g kg^{-1}	10.6	K_L**	9.4
	leaf P, g kg^{-1}	1.9	NS	14.5
		Bebedouro		
V/RL	canopy diameter, cm	276	NP*	5.1
	canopy height, cm	253	NS	5.8
	canopy volume, m^3	10.4	NS	13.7
	fruit yield, t ha^{-1}	41.0	NS	18.3
	leaf N, g kg^{-1}	29.9	N_L**	2.7
	leaf K, g kg^{-1}	15.3	K_L**	6.8
	leaf P, g kg^{-1}	1.6	P_L*	17.5
V/CL	canopy diameter, cm	297	N_L*, P_L*	5.0
	canopy height, cm	276	NS	5.4
	canopy volume, m^3	13.4	NS	12.8
	fruit yield, t ha^{-1}	26.3	N_L**, P_L*	25.9
	leaf N, g kg^{-1}	27.2	N_L**, N_Q*	2.7
	leaf K, g kg^{-1}	13.1	K_L**	8.1
	leaf P, g kg^{-1}	2.3	N_L*, P_L*	8.6

Table 2.7. Continued.

Tree †	Dependent variable	Mean ‡	F test §	CV, %
		Matão		
N/RL	canopy diameter, cm	328	NS	5.6
	canopy height, cm	307	N_L*	5.0
	canopy volume, m^3	19.0	N_L*, NP*	12.0
	fruit yield, t ha^{-1}	69.2	P_L*	16.3
	leaf N, g kg^{-1}	30.1	N_L*, PK*	2.7
	leaf K, g kg^{-1}	17.3	K_L**, P_Q*	6.5
	leaf P, g kg^{-1}	1.8	NS	17.1
N/CL	canopy diameter, cm	314	NS	8.2
	canopy height, cm	293	PK*	5.0
	canopy volume, m^3	16.7	NS	17.8
	fruit yield, t ha^{-1}	45.0	P_Q*, NK*	15.4
	leaf N, g kg^{-1}	27.6	N_L**, P_L**	2.7
	leaf K, g kg^{-1}	13.4	N_L*, P_Q*	10.6
	leaf P, g kg^{-1}	1.7	N_L**, P_L**	7.0
N/SW	canopy diameter, cm	256	NP*	8.6
	canopy height, cm	252	N_L*, P_Q*	7.4
	canopy volume, m^3	9.5	N_L*, NP*	18.6
	fruit yield, t ha^{-1}	33.2	N_Q*, K_Q*, NK**	17.8
	leaf N, g kg^{-1}	30.6	N_L**	3.2
	leaf K, g kg^{-1}	17.0	N_L*, K_L**	7.3
	leaf P, g kg^{-1}	1.8	NS	10.1

† P = 'Pera', V = 'Valencia', and N = 'Natal' sweet oranges; RL = Rangpur lime; CL = Cleopatra mandarin; and SW = Swingle citrumelo rootstocks.

‡ Fruit yield = cumulative value for the 4^{th} and 5^{th} years after tree planting ($n = 32$); other variables = average of two years of observation during the referred period ($n = 32$).

§ Subscript L or Q = linear or quadratic component of the prediction model, respectively.

NS, *, ** Nonsignificant or significant at P = 0.05 or 0.01, respectively.

Table 2.8. Response functions for citrus trees as in regard to fruit yield, canopy growth, or leaf nutrient content dependent variables in Sta. Cruz R. Pardo.

Tree †	Model coefficients ‡										R^2
	B_0	N	N^2	P	P^2	K	K^2	NP	NK	PK	
					Canopy Diameter, cm						
P/RL	Y = 327.3	-0.03790	1.50E-05	0.01339	-1.70E-06	0.02537	-4.90E-06	-3.80E-06	9.00E-07	-7.50E-06	0.45
					Canopy height, cm						
P/RL	Y = 297.0	-0.02976	9.70E-06	0.01434	-4.40E-06	0.01052	-5.20E-06	-3.40E-06	8.30E-06	-2.00E-07	0.52
					Canopy volume, m^3						
P/RL	Y = 17.3	-0.00584	2.19E-06	0.00252	-5.40E-07	0.00323	-8.80E-07	-6.70E-07	5.90E-07	-7.10E-07	0.54
					Fruit yield, t ha^{-1}						
P/RL	Y = 38.1	-0.00290	5.20E-06	0.01342	2.13E-06	0.03379	-7.26E-06	-9.02E-06	-2.30E-07	-5.98E-06	0.74
					Leaf K, g kg^{-1} §						
P/RL	Y = 9.9	-0.00152	-4.80E-08	-0.00301	5.77E-07	0.00439	-1.14E-06	7.15E-07	1.93E-07	6.96E-07	0.84

† P = 'Pera' sweet orange; RL = Rangpur lime rootstock.

‡ Rates of NPK are in g N, g P_2O_5, or g K_2O per tree. Fruit yield = cumulative value for the 4th and 5th years after tree planting (n = 32); other variables = average for two years of observation during the referred period (n = 32).

§ Six to eight month-old leaves from fruiting terminals.

as soil acidity in Bebedouro probably limited the growth of trees in comparison with other sites. On the other hand, average canopy volume of trees on Rangpur lime or Swingle citrumelo rootstocks showed significant effects of N and P fertilization in Matão (Table 2.7). The response models found for either rootstock (Table 2.10) estimated a small positive responses to N fertilization only at P_2O_5 rates above 1600 g per tree (with K_2O = 300 g per tree). Canopy volume in the fifth year of NPK fertilization and cumulative fruit yield for individual rootstocks within experimental sites were poorly correlated (Figure 2.1). The coefficients of determination (R^2) were between 0.01 and 0.32. Nonbearing trees, 4 to 5-yr-old, require nutrients for canopy growth and increasingly, for fruit yield, which can lead to an alternate bearing behavior of these trees. Since we evaluated only two years of fruit production, a large variation is expected in our data. On the other hand, the relationship found for averages of fruit yield and canopy volume for all sites showed a better agreement between such parameters (Fig. 2.1). This is possible because an ample range of canopy size is observed among rootstock species. In general, the greater the tree, the greater the fruit yield. Obreza and Rouse (1993) also found a poor correlation (R^2 = 0.20) between canopy volume of 'Hamlin' orange trees and fruit yield at 24 and 31 months after planting. The authors explained that canopy volume may not be meaningful for young citrus trees, because canopy density can vary among trees with similar canopy volume.

Table 2.9. Response functions of citrus trees for fruit yield, canopy growth, and leaf nutrient concentrations in Bebedouro.

Tree †	Model coefficients ‡										R^2
	B_0	N	N^2	P	P^2	K	K^2	NP	NK	PK	
					Canopy Diameter, cm						
V/RL	Y = 237.7	0.02061	-2.50E-06	0.00884	4.40E-06	0.03689	-1.14E-05	-1.26E-05	-3.00E-06	-6.00E-07	0.37
V/CL	Y = 255.1	0.04300	-9.50E-06	0.00223	3.70E-06	-0.00093	5.60E-06	-3.80E-06	-4.80E-06	1.30E-06	0.38
					Fruit yield, t ha^{-1}						
V/CL	Y = 18.7	0.00760	1.29E-06	-0.00386	3.76E-06	-0.01388	6.36E-06	-4.06E-06	-1.20E-07	3.30E-06	0.52
					Leaf N, g kg^{-1} §						
V/RL	Y = 28.2	0.00161	-4.90E-07	-0.00014	-2.70E-07	0.00108	-9.10E-07	1.80E-07	4.30E-07	5.30E-07	0.59
V/CL	Y = 26.4	0.00299	-1.22E-06	-0.00064	2.00E-08	-0.00067	-1.20E-07	2.20E-07	5.40E-07	2.00E-08	0.59
					Leaf P, g kg^{-1} §						
V/RL	Y = 0.7	0.00005	1.82E-07	0.00123	-2.34E-07	0.00122	-2.38E-07	-1.10E-07	-2.72E-07	-2.05E-07	0.39
V/CL	Y = 1.5	0.00012	-5.20E-08	0.00017	-3.50E-08	0.00009	-2.50E-08	-4.20E-08	-1.00E-08	-3.30E-08	0.25
					Leaf K, g kg^{-1} §						
V/RL	Y = 11.7	0.00034	-1.00E-08	0.00047	3.00E-08	0.00465	-7.60E-07	6.00E-08	-3.00E-07	-6.60E-07	0.60
V/CL	Y = 11.3	-0.00253	8.90E-07	0.00083	-8.00E-08	0.00379	-2.90E-07	1.60E-07	-4.70E-07	-6.30E-07	0.62

† V = 'Valencia' sweet orange; RL = Rangpur lime, and CL = Cleopatra mandarin rootstocks.

‡ Rates of NPK are in g N per tree, g P_2O_5 per tree, or g K_2O per tree. Fruit yield = cumulative value for the 4th and 5th years after tree planting (n = 32); other variables = average for two years of observation during the referred period (n = 32).

§ Six to eight month-old leaves from fruiting terminals.

Table 2.10. Response functions of citrus trees for fruit yield, canopy growth, and leaf nutrient concentrations in Matão.

Tree †	Model coefficients ‡										R^2
	B_0	N	N^2	P	P^2	K	K^2	NP	NK	PK	
					Canopy diameter, cm						
N/SW	Y = 278.4	0.01702	-1.79E-05	-0.00533	-1.52E-05	-0.01568	8.00E-07	1.82E-05	-2.90E-06	1.63E-05	0.40
					Canopy height, cm						
N/RL	Y = 337.3	-0.01200	1.00E-08	-0.04537	1.36E-05	0.01097	-6.00E-07	7.80E-06	-8.00E-06	1.30E-06	0.37
N/CL	Y = 273.7	-0.02731	8.10E-06	0.04477	-1.31E-05	0.01936	1.00E-09	2.40E-06	2.80E-06	-1.52E-05	0.34
N/SW	Y = 280.1	-0.03673	5.00E-07	0.02405	-2.11E-05	-0.00039	-2.90E-06	1.34E-05	6.10E-06	4.50E-06	0.47
					Canopy volume, m^3						
N/RL	Y = 23.6	0.00055	-1.36E-06	-0.00715	1.36E-06	0.00209	-7.90E-07	1.90E-06	-7.80E-07	6.30E-07	0.39
N/SW	Y = 11.4	0.00004	-1.09E-06	0.00070	-1.61E-06	-0.00080	5.00E-08	1.40E-06	-1.00E-07	1.08E-06	0.46
					Fruit yield, t ha^{-1}						
N/RL	Y = 57.8	-0.00644	1.51E-06	-0.00001	1.11E-06	0.02209	-2.85E-06	6.57E-06	-5.95E-06	-5.83E-06	0.27
N/CL	Y = 31.1	0.00712	-2.41E-06	0.01991	-8.96E-06	-0.00421	9.45E-06	5.28E-06	-6.65E-06	-4.20E-06	0.49
N/SW	Y = 10.0	0.03245	-7.95E-06	0.00061	3.47E-07	0.00355	8.28E-06	1.11E-07	-1.20E-05	-1.96E-06	0.60

Table 2.10. Continued.

Tree †	Model coefficients ‡										R^2
	B_0	N	N^2	P	P^2	K	K^2	NP	NK	PK	
					Leaf N, g kg^{-1} §						
N/RL	Y = 33.4	-0.00174	6.60E-07	-0.00214	4.20E-07	-0.00296	3.00E-07	-2.40E-07	7.50E-07	1.15E-06	0.48
N/CL	Y = 26.2	0.00057	-1.40E-07	0.00001	3.50E-07	0.00177	-3.20E-07	-1.70E-07	-5.50E-07	-1.20E-07	0.50
N/SW	Y = 28.1	0.00293	-4.30E-07	-0.00039	2.50E-07	0.00050	-3.40E-07	-4.90E-07	-3.00E-07	5.40E-07	0.43
					Leaf K, g kg^{-1} §						
N/RL	Y = 11.3	0.00348	-8.00E-07	0.00365	-1.23E-06	0.00315	-5.70E-07	-6.40E-07	-5.40E-07	3.60E-07	0.61
N/CL	Y = 13.7	-0.00138	2.80E-07	0.00398	-1.61E-06	-0.00069	-7.00E-08	-3.40E-07	2.40E-07	2.00E-07	0.37
N/SW	Y = 13.5	0.00195	-1.03E-06	-0.00056	2.20E-07	0.00525	-1.26E-06	-1.00E-08	-1.00E-08	-1.50E-07	0.68
					Leaf P, g kg^{-1} §						
N/CL	Y = 1.2	0.00033	-6.10E-08	0.00013	-9.00E-09	0.00019	-6.30E-08	1.00E-09	-1.80E-08	-1.80E-08	0.60

† N = 'Natal' sweet orange; RL = Rangpur lime, CL = Cleopatra mandarin, and SW = Swingle citrumelo rootstocks.

‡ Rates of NPK are in g N per tree, g P_2O_5 per tree, or g K_2O per tree. Fruit yield = cumulative value for the 4^{th} and 5^{th} years after tree planting (n = 32); other variables = average for two years of observation during the referred period (n = 32).

§ Six to eight month-old leaves from fruiting terminals.

Fruit Yield and Leaf Analysis

A marked response on fruit yield of 'Pera' on Rangpur lime due to the NPK fertilization was observed in Sta. Cruz R. Pardo (Table 2.7). The response model estimated about 50% fruit yield increase with an increase in K rates from 300 to 1800 g K_2O per tree (with 400 g of N and P_2O_5 per tree) (Table 2.8). Current recommended rates of K fertilizer for nonbearing citrus trees in Brazil sum up to 750 g K_2O per tree during five years after tree planting (Grupo Paulista, 1994). Average K concentration in the leaves from fruiting terminals also increased by K fertilization (Table 2.7), which in turn was well correlated with soil exchangeable K (Fig. 2.2). The relationship between K and citrus production is reported to be nonsignificant in Florida (Hunziker, 1960; Koo, 1962; Hanlon et al., 1995). On the other hand, studies conducted in São Paulo, Brazil, have shown significant relationship between fruit yield of 'Pera' and lemon trees, and exchangeable K levels following various levels of K fertilization (Cantarella et al., 1992; Quaggio et al., 1996). Maximum fruit yield of 'Pera' on Rangpur lime trees was observed with soil exchangeable K level about 2.0 $mmol_c\ dm^{-3}$ (Figure 2.2). The critical level of K in the soil as related to fruit yield reported for bearing trees in Brazil is 2.0 $mmol_c\ dm^{-3}$ (Quaggio et al., 1996). Average cumulative fruit yield increased about 35 $t\ ha^{-1}$ as soil pH increased up to 6 (Fig. 2.3). Such observation is related to the acidification of the soil surface layer caused by the addition of N fertilizer, and explains the negative linear term of the response model for the N effect in Sta. Cruz R. Pardo (Table 2.8).

The lack of significant response of fruit yield to P fertilization in Sta. Cruz R. Pardo (Table 2.7) can be explained by the fact P was mechanically incorporated in three furrows along the tree row before planting. Panzenhagen et al. (1999) reported that fruit

yield of 'Montenegrina' tangerines was unaffected by application of P until 8 years because of the corrective fertilization before tree planting. Furthermore, initial P level at soil surface was relatively high (22 mg dm^{-3}; Table 2.2) and increased to above 60 mg dm^{-3} in the fifth year of P fertilizer application (Table 2.5). The significant NP interaction for fruit yield (Table 2.7) shows an antagonism between those two nutrients at high N rates (Table 2.11). Wallace (1990) reported a severe antagonism for N and P fertilization of 'Valencia' orange trees, which resulted in a large fruit yield decrease. Davies and Albrigo (1994) also reported that N and P leaf levels are inversely related.

Cumulative fruit yield of 'Valencia' orange trees on Rangpur lime rootstock was not affected by the NPK fertilization in Bebedouro (Table 2.7), although average cumulative production was about 35 t ha^{-1} (Fig. 2.4A). Trees on Cleopatra mandarin showed significant responses to N and P fertilization (Table 2.7). The yield increased from 17 to 34 t ha^{-1} as the N rate increased from 400 or 2200 g per tree at 400 and 300 g of P_2O_5 or K_2O per tree, respectively (Table 2.9; Fig. 2.4A). However, leaf analysis showed that the N concentration was greater for trees on the Rangpur lime (29.6 g N kg^{-1}) as compared to those on Cleopatra mandarin (27.5 g N kg^{-1}) across all N rates (Fig. 2.5A). Reported values for N concentration in the leaves of these trees were greater than the adequate levels for bearing trees (23-27 g kg^{-1}) as reported by Grupo Paulista (1994). Trees on Cleopatra mandarin were vigorous with larger canopy as compared to those on Rangpur lime rootstock. Therefore the lower N concentration in the leaves of the former can be attributed to a dilution effect of the nutrient in the tree biomass (Wutscher, 1989). The response of fruit yield and leaf analysis to P fertilization was also very distinct for trees on different rootstocks (Table 2.7). Fruit yield of the trees

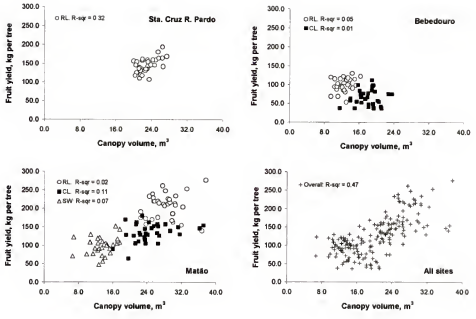

Figure 2.1. Relationship between cumulative orange fruit yield and canopy volume at different locations.

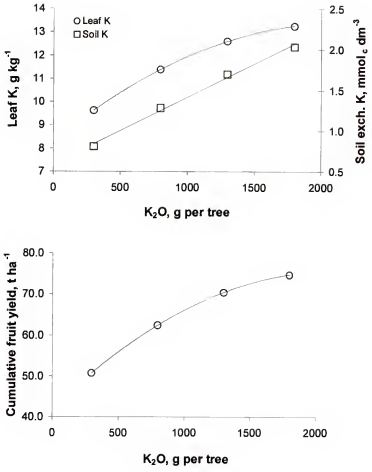

Figure 2.2. Effects of K fertilization on concentration of leaf K, soil exchangeable K, and cumulative fruit yield of 'Pera' sweet orange on citrumelo Swingle rootstock in Sta. Cruz R. Pardo as estimated by response models (with N and P = 400 g of N or P_2O_5 per tree, respectively).

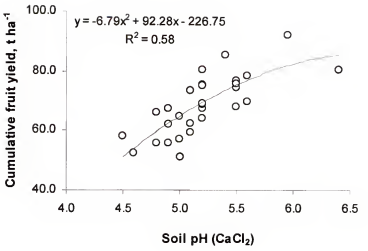

Figure 2.3. Effect of soil pH on fruit yield of 'Pera' sweet orange on Rangpur lime rootstock in Sta. Cruz R. Pardo.

on Cleopatra mandarin increased significantly from 17 and 28 t ha^{-1} with an increase in P_2O_5 rate from 400 to 2200 g per tree at 400 g of N and 300 g K_2O per tree (Fig. 2.4B). The concentration of P in the leaves of the trees on Rangpur lime was about 1.6 g kg^{-1} and did not change much with P fertilization, whereas that of the trees on Cleopatra mandarin increased from 1.4 to 2.3 g kg^{-1} with an increase in P_2O_5 from 400 to 2200 g per tree (Fig. 2.5B). Samiullah and Narasimham (1979) reported the leaf P concentration of a sweet orange scion was lowest for the trees on Cleopatra mandarin rootstock as compared to that of the trees on citrange Troyer, Volkamer lemon, or Rough lemon rootstocks. Wutscher (1989) also reported low concentration of N and P in the leaves of several sweet orange cultivars on Cleopatra mandarin as compared to other rootstocks, while the concentration were greater for the trees on Rangpur lime. Figure 2.6 shows the

Table 2.11. Estimated cumulative fruit yield for 'Pera' orange trees on Rangpur lime rootstock as affected by the N and P fertilization (with K = 1800 g K_2O per tree).

N, g per tree	P_2O_5, g per tree			
	400	1000	1600	2200
	Cumulative fruit yield, t ha^{-1}			
400	74.9	76.1	78.9	83.2
1000	75.1	73.1	72.6	73.6
1600	79.1	73.8	70.0	67.8
2200	86.8	78.2	71.3	65.8

relationship between P fertilization and P concentrations in the soil (resin extracted) and in the leaves of 'Valencia' orange trees on Cleopatra mandarin. Fruit yield increased above critical levels for soil P (20 mg dm^{-3}) established for bearing trees (Quaggio et al., 1996). Similarly, leaf P concentration was greater than adequate levels for bearing trees (1.2-1.6 g kg^{-1}) as suggested by Grupo Paulista (1996). Nonbearing trees have a smaller root system as compared to that of the mature trees, therefore, the former could respond to higher concentrations of soil P since the volume of soil explored by roots is much smaller and the band where fertilizer is applied to the soil surface changes annually as the canopy diameter increases.

There was no significant response of fruit yield to K fertilization in Bebedouro (Table 2.7), even though concentration of total K in the leaves increased with K rates (Fig. 2.5C). Soil exchangeable K was up to 4.2 $mmol_c$ dm^{-3} in the 5th year of treatment application (Table 2.5). The lack of fruit yield response with increasing levels of leaf K can be attributed to luxurious absorption of K (Marschner, 1995).

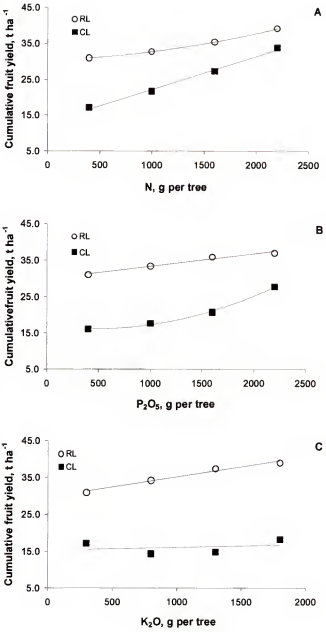

Figure 2.4. Effects of the NPK fertilization on cumulative fruit yield of 'Valencia' sweet orange trees on Rangpur lime (RL) or Cleopatra mandarin (CL) rootstocks in Bebedouro as estimated by response models (with N and P = 400 g of N or P_2O_5 per tree, respectively, and K = 300 g K_2O per tree). a) Response to N; b) Response to P; c) Response to K fertilization.

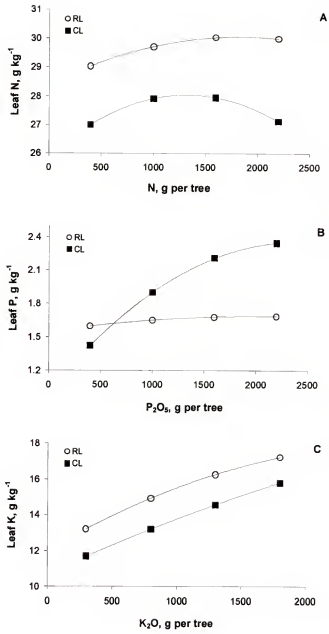

Figure 2.5. Effects of the NPK fertilization on the leaf concentration of nutrients of 'Valencia' sweet orange trees on Rangpur lime (RL) or Cleopatra mandarin (CL) rootstocks in Bebedouro as estimated by response models (with N and P = 400 g of N or P_2O_5 per tree, respectively, and K = 300 g K_2O per tree). a) Response to N; b) Response to P; c) Response to K fertilization.

Distinct effects of NPK fertilization on fruit yield and leaf analysis were evident in Matão (Table 2.7 and 2.10). 'Natal' sweet orange trees on Swingle citrumelo rootstock showed a more pronounced response of cumulative fruit yield to N fertilization (up to 38 t ha^{-1}, with P = 400 g P_2O_5 and K_2O = 300 g K_2O per tree) (Fig. 2.7A). The leaf N concentrations of these trees increased from 29 to 32 g kg^{-1} with an increase in N rate from 400 to 2200 g per tree (Fig. 2.8A). On the other hand, the N concentration in the leaves of the trees on either Rangpur lime or Cleopatra mandarin did not change with N fertilization, and were about 31 and 26 g kg^{-1}, respectively (Fig 2.8A). Positive effects of P fertilization on fruit yield and leaf P concentration were evident only for trees on Cleopatra mandarin rootstock. Cumulative fruit yield increased from 40 to 56 t ha^{-1} with an increase in P_2O_5 rates from 400 to 2200 g per tree (with N = 400 g of N, and K = 300 g K_2O per tree) (Fig. 2.7B). The corresponding concentrations of P in the leaves increased from 1.5 and 1.8 g kg^{-1} (Fig. 2.8B). This range is above the adequate concentration for bearing trees reported by Grupo Paulista (1994). In the case of the trees on Rangpur lime or Swingle citrumelo rootstocks P fertilization effects were nonsignificant on leaf P concentration (Fig. 2.8B). The increase in fruit yield for K fertilization was marked for the trees on Swingle citrumelo as compared to that of the trees on the other two rootstocks (Table 2.2). Cumulative fruit yield ranged from 22 to 43 t ha^{-1} at K rates of 300 or 1800 g K_2O per tree, respectively (with N and P = 400 g of N or P_2O_5 per tree) (Fig. 2.7C). The leaf K concentrations increased from 15 to 19 g kg^{-1} (Fig. 2.8C). The above range of leaf K concentrations are greater than those reported as the adequate range for bearing trees (10-15 g kg^{-1}; Grupo Paulista, 1994).

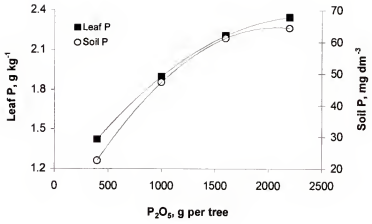

Figure 2.6. Correlation between P fertilization and soil available P or leaf P concentrations for 'Valencia' sweet orange trees on Cleopatra mandarin rootstock in Bebedouro as estimated by response models (with N = 400 g N and K = 300 g K_2O per tree).

Our findings are in agreement with data reported by Wutscher (1989), which suggest that leaf K concentrations are greater for the trees on Swingle citrumelo rootstock compared to that of the trees on other rootstocks. Responses of trees on Cleopatra mandarin was in between Swingle citrumelo and Rangpur lime rootstocks. The significant N interaction with K found for fruit yield of trees on Swingle citrumelo is showed in Table 2.12. Larger fruit yield was obtained with low rates of N and high rates of K (45.2 t ha^{-1}) or inversely, with low rates of K and high rates of N (36.7 t ha^{-1}). DuPlessis and Koen (1988) reported an antagonistic effect of N and K fertilization on fruit yield and size of 'Valencia' orange trees. In this experiment, high K applications only increased the leaf K concentration if relatively low N rates were applied. In general, N and K concentrations in leaves are inversely related (Davies and Albrigo, 1994).

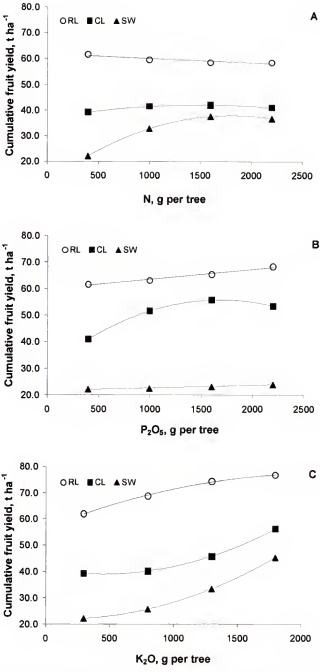

Figure 2.7. Effects of the NPK fertilization on cumulative fruit yield of 'Natal' sweet orange trees on Rangpur lime (RL), Cleopatra mandarin (CL) or Swingle citrumelo (SW) rootstocks in Matão as estimated by response models (with N and P = 400 g of N or P_2O_5 per tree, respectively, and K = 300 g K_2O per tree; in the case of trees on CL, the response to P fertilization was estimated with N = 2200 g per tree). a) Response to N; b) Response to P; c) Response to K fertilization.

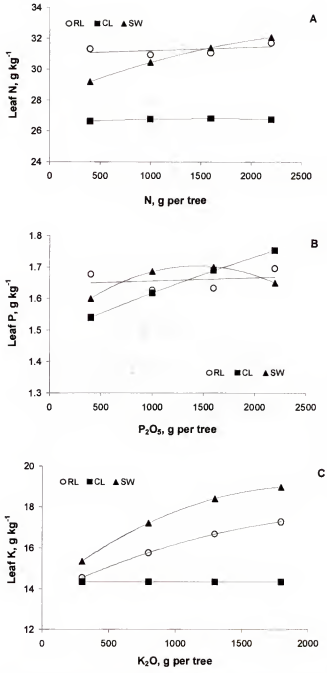

Figure 2.8. Effects of the NPK fertilization on cumulative fruit yield of 'Natal' sweet orange trees on Rangpur lime (RL), Cleopatra mandarin (CL) or Swingle citrumelo (SW) rootstocks in Matão as estimated by response models (with N and P = 400 g of N or P_2O_5 per tree, respectively, and K = 300 g K_2O per tree). a) Response to N; b) Response to P; c) Response to K fertilization.

Table 2.12. Estimated cumulative fruit yield for 'Natal' orange trees on Swingle citrumelo rootstock as affected by the N and K fertilization (with P = 2200 g P_2O_5 per tree).

N, g per tree	K_2O, g per tree			
	300	800	1300	1800
	Cumulative fruit yield, t ha^{-1}			
400	22.2	25.7	33.4	45.2
1000	32.8	32.7	36.8	45.1
1600	37.6	34.0	34.5	39.1
2200	36.7	29.5	26.4	27.4

Current recommended rates of N and P fertilizers for nonbearing citrus trees in Brazil sum up to 1140 g N and 1060 g P_2O_5 per tree during five years after tree planting (Grupo Paulista, 1994). Estimated data showed that fruit yield of orange trees can increase with rates higher than those recommended depending on the rootstock (i.e., Cleopatra mandarin is more responsive to P fertilization). Leaf nutrient analysis showed that the critical limits of leaf concentration of N and P for nonbearing trees could be greater than those reported by Grupo Paulista (1994) for bearing trees. Willis et al. (1990), and Obreza and Rouse (1993) found that leaf concentration of selected nutrients for young trees tended to be greater than those recommended for bearing trees in Florida (Koo et al., 1984).

Conclusion

Rootstocks influence the canopy size of citrus trees, as well as the fruit yield response to NPK fertilization. Linear responses of fruit yield to N fertilization were obtained for the trees on Cleopatra mandarin rootstock with an increase in N rate from 400 to 2200 g N per tree (total for 5 years) in Bebedouro and Matão, while fruit yield of the trees on Swingle citrumelo responded to a lesser extent depending on K rates. Fruit yield also increased with P_2O_5 rates up to 2200 g per tree, for the trees on Cleopatra mandarin rootstock only. Increased concentrations of leaf P correlated with the increase in fruit yield. A marked linear response to the K fertilization was observed up to 1800 g K_2O per tree (total for 5 years) in Sta. Cruz R. Pardo, where the soil exchangeable K was low. Fruit yields were well correlated with leaf K concentrations or the soil exchangeable K levels. Trees on Swingle citrumelo in Matão also responded linearly to K rates. Our findings suggest that rates of NPK fertilizers to maximum growth and production of nonbearing trees depend on the rootstocks. Further, the critical ranges of leaf nutrient concentrations for growth and specially fruit yield of nonbearing were higher than those reported for the bearing trees. The data from this study also suggested acidification of surface soil by increased N rates, which in turn affected the tree growth and consequently fruit yield. The soil pH about 6 supported maximum fruit yield.

CHAPTER 3
VOLATILIZATION OF AMMONIA FROM N FERTILIZERS APPLIED TO THE SOIL SURFACE OF A CITRUS GROVE

Introduction

Urea is the most common nitrogen fertilizer used for various crops around the world. Its widespread use is due to its low cost per unit of N and the high content of nitrogen (N) which is favorable for handling, storage, transportation (Havlin et al., 1999) and application in the field. However, N uptake efficiency from urea is quite low if it is not incorporated, because of volatilization of ammonia (NH_3). Gaseous loss from soil following urea fertilizer application may account for up to 75% of total N applied (Fenn and Miyamoto, 1981), otherwise volatilization from other forms of nitrogen fertilizer is less significant than from urea in acid soils (Havlin et al., 1999).

Volatilization loss of NH_3 is a result of biochemical reactions in the soil. The process of N volatilization from applied urea is related to a localized increase in soil pH after dissolution and hydrolysis of the fertilizer (Hauck, 1984). Heterothrophic microbes are responsible in part for the ammonification of urea-amino group, which results in the production of the ammoniacal compound, which is then released into the soil solution (Myrold, 1998). The hydrolysis of urea is a process catalyzed by enzymes of the aminohydrolases group present in the soil, of which urease is one of the most important extracellular enzymes (Tabatabai, 1994). Its presence in soil is related to a large number of bacteria, fungi and actnomycetes (Myrold, 1998). Although urease is common in soils,

its activity is a function of many soil properties, such as pH, temperature, moisture, texture, buffer capacity and organic C content (Bremner and Mulvaney, 1978). The greatest activity of urease is in the rhizosphere, where microbial activity is high and organic carbon is available as plant root exudates. High temperature also enhances the activity of such enzymes up to the limit of 70°C (Voss, 1984).

Studies on N volatilization have been carried out in the laboratory, greenhouse, and in the field. However, within restricted environments, the soil-atmosphere conditions may differ greatly from those in the field. Hargrove and Kissel (1979) found that in field conditions, NH_3 losses from urea fertilizer applied to Coastal bermudagrass sod were lower (0 to 9%) than those in laboratory conditions (13 to 31%). Freney et al. (1992) observed significant ammonia losses from urea applied to sugarcane in Australia over a period of six weeks. In a dry climatic zone ammonia loss was 39% of the applied N but decreased to 17% in a wet zone. The rate and extent of ammonia losses reported in this work were controlled by the availability of water in the soil system. Alternate wetting and drying cycles accelerated the volatilization loss. Ammonia losses were quite low, i.e. 1.8% of the applied N from $(NH_4)_2SO_4$. Surface applied urea to a permanent orchardgrass sod resulted in NH_3 losses up to 42% within 8 days (Lightener et al., 1990). The losses were lower under extremely dry conditions. Soil cation exchange capacity (CEC) also affects NH_3 volatilization loss. Keller and Mengel (1986) reported that total gaseous losses of N in 5 days from surface applied urea to no-till corn were 30 and 11% from a Mollisol with CEC of 6.7 $cmol_c\ kg^{-1}$, and an Alfisol with CEC of 12.3 units. Both soils had similar pH.

Ammonia losses from urea are greater when applied to a soil surface covered with plant material. Urban et al. (1987) found that NH_3 losses from urea measured in growth chamber experiments ranged from 15 to 32% of the applied N on a bare soil, and 68 to 82% on a mulched soil. The effects of plant residue mulch on NH_3 volatilization were due to (i) a physical barrier between the N source and the soil by the crop residue, (ii) by maintaining the soil in moist condition over a prolonged period, (iii) a relatively high urease activity of the plant residue. Therefore, it appears that the potential for NH_3 losses is greater when urea is applied to surface soils under no-tillage management. Citrus orchard management is changing to minimize weed competition and enhance early fruit production for maximum economic returns. Herbicide is commonly applied in a band of about 1.5 m on both sides of the tree row, and the grass in the middle row mowed periodically is mechanically spread under the tree canopy, where the fertilizer is generally broadcast. The increased plant residue on the soil surface could influence the volatilization loss. Incorporation of fertilizers with soil in citrus groves is often difficult due to the problem of plant injury. Broadcast application of urea is most common which can lead to substantial losses of N due to NH_3 volatilization (Cantarella and Quaggio, 1996).

There are different types of devices used to measure N gaseous losses from soils (Marshall and Debell, 1980). It is common to use acid sorbers for trapping ammonia, even though methods differ in the mechanism by which NH_3 and other gases may move from the soil to the sorber. Nômmik (1973a) used a semi-open system, which permits exchange with the atmosphere via diffusion. Kissel et al. (1977), Ferguson et al. (1988) and Sherlock et al. (1989) tested variations of this semi-open system in the field, which

allowed continuous measurement of NH_3 volatilization over a broad range of environmental conditions and with a number of different treatments. In a semi-open environment, the diffusion rate of NH_3 can differ from that in natural conditions. This in turn could influence the volatilization loss. Another type of measurement of NH_3 volatilization is possible by computing the vertical and horizontal fluxes of atmospheric NH_3 concentration and wind speed in large circular plots (mass balance micrometeorological method) (Wilson et al., 1982, 1983; Black et al., 1985a; Leuning et al., 1985; Wood et al., 2000). In this case, there is no disturbance of atmospheric conditions near the soil surface. This method requires extensive site instrumentation. Its applicability to areas grown with tree species is more difficult, because of the non-uniform environment, especially wind movement, created by the disposition of tree rows and inter-rows as compared to pasture areas.

The field evaluation of NH_3 losses is important to estimate the efficiency of applied nitrogen fertilizer in order to develop estimates of N budget for a given cropping system. The objective of this research was to determine the NH_3 volatilization losses from urea or NH_4NO_3 surface applied to an Alfisol under 6-yr-old citrus.

Material and Methods

Characteristics of the Study Area and Experimental Design

A field experiment was conducted to study NH_3 volatilization in a commercial citrus grove of 'Valencia' sweet orange trees (*Citrus sinensis* (L) Osb.) on Rangpur lime rootstock (*C. limonia* Osb.) established in an Alfisol of the São Paulo State, Brazil, in 1990. Soil water storage determined as the difference of precipitation and evapotranspiration for the area is shown in Fig. 3.1. The rainy season usually ends after May or June, even though dry periods may start earlier. Average maximum and minimum air temperatures recorded since 1950 are 28.6 and 17.2°C in April and 26.2 and 14.7°C in May, respectively. Selected soil physical and chemical characteristics are presented in Table 3.1. The trees were planted in a 7.5 x 4.2 m spacing. Weed control was done by applying herbicide in a band of 1.5 to 2.0 m wide on both sides of the tree row only; the grass in the middle was mechanically mowed periodically during the course of the growing season. Weed population on the citrus grove was represented mostly by kikuyugrass (*Pennisetum clandestinum* Hochst.), digitgrass (*Digitaria insularis* (L.) Mez. ex Ekman), and hairy beggarticks (*Bidens pilosa* L.). The NH_3 volatilization experiment started in 1997 had a randomized block design with treatments represented by urea (45%N) or NH_4NO_3 (33%N) fertilization, which were replicated 4 times. The annual N rates used were 20, 100 and 260 kg N ha^{-1} for urea, and 20 and 260 kg N ha^{-1} for ammonium nitrate, applied in equal doses in October, January and April. The fertilizer was hand applied on the soil surface, in 1.5 m wide bands of on both sides of the tree line. About 65% of the fertilizer was applied under the tree canopy.

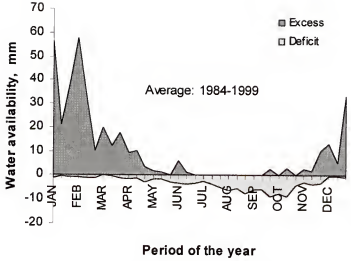

Figure 3.1. Soil water storage determined as the difference between precipitation and evapotranspiration through the year. Matão, 1984-1999.

A semi-open system of NH_3 sorbers was used to evaluate the gaseous loss of fertilizer N, following the model described by Nômmik (1973a). The trapping device consisted of a PVC-cylinder, 35-cm long and 20-cm in diameter. Two discs of polyurethane plastic foam (density: 0.03 g cm^{-3}; thickness: 2 cm; and circular area: 314 cm^2) soaked in 2.25 mol L^{-1} H_3PO_4 plus 4.0% (v/v) glycerol solution were placed inside the PVC-cylinder at 10 and 20 cm from the top. Each disc retained 100 to 120 mL of the acid-glycerol solution. The disc placed at 10 cm from the top of the cylinder was used to trap potential contamination of the atmospheric NH_3. The lower position disc traps the NH_3 volatilized from the soil and was used to measure the losses.

Collectors were placed on the soil surface within the fertilized area close to the tree canopy immediately after the third dose of annual N on April 9, 1996, April 16,

Table 3.1. Selected soil physical and chemical characteristics.

Depth	OM	pH †	Ca	CEC	BS ‡	Clay	Sand
cm	g kg^{-1}		----- $mmol_c$ kg^{-1} -----		%	---------- % ----------	
0-20	20	5.4	29.3	66.2	68.6	13.1	80.4
20-40	12	5.4	24.8	58.0	70.3	16.6	76.9
40-60	9	5.4	23.5	60.2	68.8	22.6	72.6

† Soil / $CaCl_2$ 0.01 mol L^{-1}, 1:2.5 ratio (v/v).
‡ Soil base saturation.

1997, and April 20, 1999. Following 2 to 5-day exposure after fertilization, the NH_3 sorber discs were removed and replaced by new treated discs. The collectors were relocated to adjacent fertilized areas following replacement of exposed discs. Discs collected in the field were placed in sealed plastic bags, taken to the laboratory, and stored in a refrigerator at 5°C. Ammonia trapped by the plastic foam discs at the lower position in the PVC-cylinder was extracted by leaching with 100 mL 1.0 mol L^{-1} KCl solution. The KCl was extracted from the foam by squeezing. This procedure was repeated 5 times and the KCl was pooled. An aliquot of KCl extract was steam-distilled with 10 mL 5.0 mol L^{-1} NaOH solution, and then titrated with H_2SO_4 0.025 mol L^{-1} for NH_4-N determination (Bremner, 1965).

The effectiveness of the upper position disc to prevent the NH_3 contamination from the external air was evaluated in 1997. The NH_3 trapped by the foam disc after application of urea (UR) or ammonium nitrate (AN) fertilizers was measured from April 16 to 29, under the following conditions: (i) UR applied to the soil outside and inside the collector; (ii) AN applied outside and inside system; (iii) UR applied outside and AN applied inside system; and (iv) AN applied inside and UR applied outside system. The

discs were replaced three times during the evaluation interval. A set of disc was also hung in the open under the tree canopy to measure NH_3 volatilization after N fertilization with either UR or AN in the citrus grove. The foam discs hung from the trees should be capable of trapping this ammonia

Data Analysis

Data were tested for significant differences among treatments in a randomized block ANOVA with four replications using the GLM procedure of the SAS® system (SAS Institute, 1996). Regression analysis was performed with data collected for NH_3 volatilization from fertilized plots using the SAS REG procedure. The model was of the type $y = a + b/x$; where y = cumulative NH_3-N loss (% of applied N), and x = time interval after fertilization (day).

Results and Discussion

Ammonia Volatilization from Soil Applied N Fertilizers

There was a good correlation between the cumulative amount of NH_3 volatilized and the time of measurement through the three years of evaluation as indicated by R^2 values ranging from 0.69 to 0.99 (Table 3.2). At the low N rate, the regression was nonsignificant ($P>0.06$), probably because the amount of ammonia being volatilized from soil surface was low during the entire experiment.

The peaks of NH_3 losses occurred within 3 days after fertilizer application for urea and with less intensity for NH_4NO_3 treatments (Fig. 3.2 to 3.4). Black et al. (1985a)

also reported low volatilization losses during the first 24 h due to the low hydrolysis, and that most of the volatilization losses were completed during the 6 days following fertilizer application, which accounted 24 to 30% of the total N applied. Under dry conditions, dissolution and hydrolysis of fertilizer granules are dependent on the addition of small amount of water through dew, and that the maximum daily losses occurred on the 3rd day (Black et al., 1985b; Keller and Mengel, 1986). In an experiment on orchardgrass sod with surface application of 200 kg N ha^{-1} as urea resulted in measurable NH_3 losses within 12 to 24 h, with a maximum rate at 3 days (Lightner et al., 1990). Cumulative losses of NH_3 in that experiment ranged from 10 to 40% of applied N. The kinetics of NH_3 volatilization from surface applied N fertilizers to an Alfisol (pH 7.9) in laboratory conditions followed a Langmuir kinetic model, i.e., an initial rapid reaction followed by a slow reaction (He et al., 1999).

The cumulative amount of NH_3 volatilized in the citrus grove from the unfertilized treatment was quite low during all three years. The ammonia loss from ammonium nitrate was 10% of applied N with 6.7 kg N ha^{-1} rate (0.7 kg N ha^{-1}; 1997) (Fig. 3.3), and about 4% with 86.7 kg N ha^{-1} rate (3.2 kg N ha^{-1}; 1999) (Fig. 3.4). With urea, however, cumulative ammonia losses accounted 17 to 26%, 15 to 31%, and 17 to 44% with 6.7, 33.3, and 86.7 kg N ha^{-1} rate, respectively (Fig. 3.2 to 3.4). The above results agree with those reported by Keller and Mengel (1986), Lightner et al. (1990), and Black et al. (1985b). Differences in the total ammonia losses from applied ammonium nitrate or urea are positively related to the maximum soil surface pH produced by each of the fertilizers after dissolution and hydrolysis (Black et al., 1985b; Fan and Mackenzie, 1993; Zia et al., 1999). A significant increase in soil pH is expected to occur after urea

Table 3.2. Regression coefficients of models for cumulative NH_3 volatilization in April as a function of time after fertilizer N application to the soil surface.

Treatment	Model coefficients †		R^2	P > F ‡
	a	b		
			1996	
UR 6.7	5.93	-5.67	0.87	0.0673
UR 33.3	1.79	-1.54	0.99	0.0022
UR 86.7	26.87	-9.08	0.99	0.0002
AN 6.7	31.69	-5.12	0.99	0.0014
AN 86.7	44.55	-6.95	0.99	0.0048
			1997	
UR 6.7	8.77	-17.22	0.71	0.1537
UR 33.3	2.02	-3.45	0.88	0.0579
UR 86.7	15.65	-23.66	0.83	0.0843
AN 6.7	15.21	-10.32	0.88	0.0571
AN 86.7	18.78	-21.28	0.94	0.0271
			1999	
UR 6.7	6.08	-12.37	0.69	0.0192
UR 33.3	3.63	-6.45	0.85	0.0027
UR 86.7	18.23	-23.18	0.94	0.0003
AN 6.7	25.76	-20.56	0.99	0.0001
AN 86.7	33.68	-18.71	0.99	0.0001

† $y = a + b/x$; where y = cumulative $N\text{-}NH_3$ loss (% of applied N), and x = time interval after fertilization (day).

‡ Model regression significance.

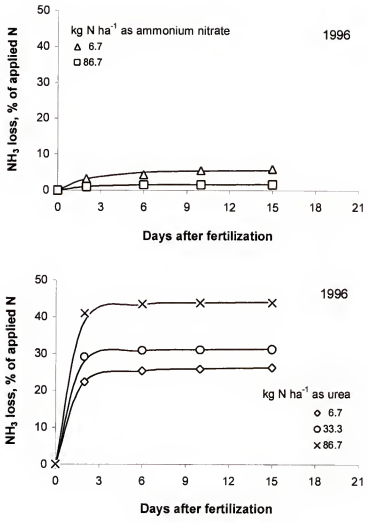

Figure 3.2. Cumulative NH_3 volatilization from soil applied N fertilizers in 1996.

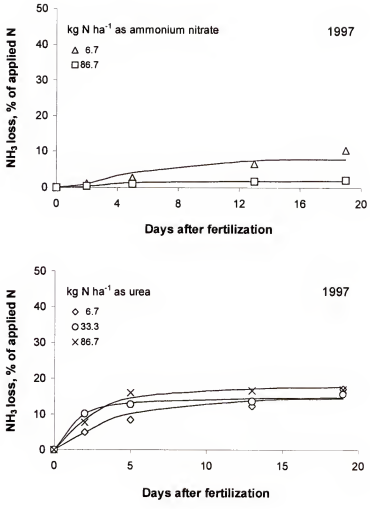

Figure 3.3. Cumulative NH_3 volatilization from soil applied N fertilizers in 1997.

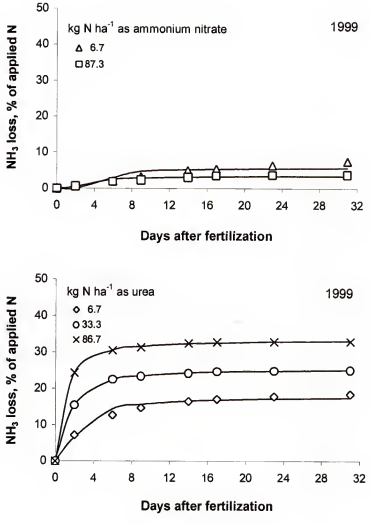

Figure 3.4. Cumulative am NH_3 volatilization from soil applied N fertilizers in 1999.

fertilization which will determine a more favorable microenvironment for NH_3 volatilization from soil surface.

The mean NH_3 volatilization losses across three years from soil applied urea were 20, 24, and 31% with application of 6.7, 33.3, and 86.7 kg N ha^{-1}, respectively. Black et al. (1985b) reported that the percentage N volatilization from urea broadcasted on to a pasture in New Zealand increased with N application rates. This was directly related to an increase in surface soil pH. Considerable overlapping of urea from adjacent granules would occur at higher rates, then surface soil pH is likely to depend in part on the degree of urea diffusing in the microsites of acid soils (Peoples et al., 1995). The weather conditions after fertilizer application influenced the N losses. Rates of ammonia volatilization were lower after fertilization in 1997 (Fig. 3.3). This was likely due to the light rain immediately after fertilization that promoted dissolution and further incorporation of fertilizer into the soil. The subsequent days were marked by a dry period that had occurred for 5 days after fertilization, when the orange grove received a 42 mm rain shower that effectively stopped volatilization. Then, between the 2^{nd} and the 5^{th} days after fertilization, the rate of urea hydrolysis in the soil probably decreased significantly until the remaining N was leached out of the surface soil by percolating water. Keller and Mengel (1986) observed that a 25 mm rain shower effectively stopped volatilization of ammonia from granular urea applied (168 kg N ha^{-1}) to a Mollisol 50 h after fertilization. A similar effect was reported by Ferguson et al. (1988); i.e., a 28 mm rain slowed the NH_3 loss from urea solution applied (120 kg N ha^{-1}) to a Mollisol. In this latter study, the cumulative losses of ammonia were less than 10% of applied N.

Lara-Cabezas et al. (1999) estimated the collector efficiency factor (E) for a semi-open static system similar to that used by Nômmik as 1 to 50%, by estimating the ratio of volatilized ammonia retained in the collector with ^{15}N mass balance method. The authors found models that would correct the lower efficiency of the semi-open system. Then, estimated losses in the present study could be lower than the real condition.

Effectiveness of the Upper Sponge Disc in the Volatilization Trap Chamber to Minimize Contamination of External Ammonia

The ammonia trapped by lower foam discs showed no significant differences ($P>0.05$) despite the N source applied to the soil outside the collector when urea or NH_4NO_3 were applied inside (Table 3.3). Greater contamination of foam discs with atmospheric ammonia would be expected with application of urea outside the collector. In this case, the NH_3-N that was found in the upper foam disc averaged 11.4 mg N (Table 3.3). Such amount is about 10 times greater than that determined when ammonium nitrate was applied outside the collector. This confirms the capability of the semi-open system to hinder cross contamination of atmospheric ammonia as reported by Nômmik (1973a).

Ammonia-N Recovered from Under the Tree Canopy Environment

The amount of ammonia trapped by the foam discs hung from the tree canopy within two days after N fertilization was 10.75 mg N per disc with urea (UR) and 2.37 mg N per disc with ammonium nitrate (AN). Leaf absorption of volatilized N can be possible at high NH_3 concentration in the air. Farquhar et al. (1980) suggested that, if the ammonia partial pressure in the leaf substomatal cavities is less than the ambient partial pressure (typically 1 to 8 nbar in unpolluted areas), there should be a net influx into the

Table 3.3. Ammonia trapped by sponge discs used in the upper and lower portions of the semi-open collector system with application of either urea (UR) or ammonium nitrate (AN).

Nitrogen source		Ammonia trapped by discs †	
Outside collector	Inside collector	Lower position	Upper position
		---------- mg NH_3-N per collector ----------	
AN	AN	3.9 b	1.6 a
AN	UR	32.0 a	1.0 a
UR	UR	52.4 a	8.0 b
UR	AN	4.7 b	14.7 b

† Same letters show no significant differences between means reported within columns at P = 0.05 significance level. Mean values (n = 4).

leaf. A mechanistic, dynamic compensation point model to simulate exchange of ammonia was suggested by Flechard et al. (1999). Ping et al. (2000) confirmed possible absorption of volatilized N by crop canopy under field conditions. In their study, wheat plants absorbed 15% of the urea-^{15}N that was volatilized after soil application at an equivalent rate of 100 kg N ha^{-1}. Therefore, actual tree recovery of soil applied N could be overestimated in specific situations due to absorption of volatilized N fertilizer.

Conclusion

Ammonia volatilization accounted for 26 to 44% of applied N as urea at 86.7 kg N ha^{-1}. The losses decreased with lower application rate. With application of NH_4NO_3, losses were 10% at 6.7 kg N ha^{-1} and 4% at 86.7 kg N ha^{-1}. Since less N was lost from NH_4NO_3, it may be a preferred N source for broadcast surface application when NH_3 volatilization is a concern. Rainfall affected the pattern of NH_3 evolution after fertilizer

application in 1997, when cumulative losses were less than 17% of applied N. The semi-open trapping device was effective in preventing contamination of the inner system with atmospheric NH_3.

CHAPTER 4
NITROGEN MINERALIZATION AND VOLATILIZATION IN A SANDY FLORIDA ENTISOL UNDER CITRUS TREES

Introduction

This research was part of an ongoing project to develop best management practices (N-BMP) for nonbearing and bearing citrus trees in Florida. This was necessary because of increasing levels of nitrate concentration in ground water near major citrus production regions in Florida. Investigations on nutrient management strategies and their impact on all components of citrus N budget in the field, including plant uptake and various losses of plant available nitrogen in the soil, which can result in adverse environmental impacts are required to address this issue in a comprehensive manner.

The mineralization of nitrogen (N) from soil organic matter, crop residues, composts, and animal manure can contribute significantly to the soil mineral-N reserve (Meisinger, 1984). The amount of net N mineralized (N-min) in soil ranges from 50 to 150 kg ha^{-1} yr^{-1} (Stanford, 1982). Studies have shown the importance of N-min for predicting N requirements of various crops under different conditions (Adams et al., 1989; Rice and Havling, 1994). Dou et al. (1997) estimated the contribution of N from mineralization of leaf residues under the citrus tree canopy as 40 to 150 kg N ha^{-1} $year^{-1}$ depending on the soil type and tree age. Dou and Alva (1998a) found a rapid increase in NH_4-N concentration in soil amended with citrus leaves during a short period of incubation in a pot experiment with Candler fine sand.

The current interest in nutrient cycling in both managed and natural ecosystems has led to renewed attempts to develop reliable methods for measuring N mineralization using either laboratory or field techniques (Rees et al., 1994). Soil N mineralization evaluation has emphasized the potentially mineralizable N (N_0) indexes as laboratory-based methods which use either aerobic (Stanford and Smith, 1972) or anaerobic (Waring and Bremner, 1964) incubation of soil samples. Keeney (1982) discussed chemical extraction procedures, based on an empirical assessment of organic-N pools. In situ methods using soil cores have been expected to give more reliable estimates of soil N mineralization than laboratory methods, because soil N turnover is strongly affected by microenvironmental conditions (Stenger et al., 1996). Soil disturbance during sample preparation markedly affects N mineralization (Nordmeyer and Richter, 1985; Kristensen et al., 2000). In the absence of plant uptake, denitrification, and leaching of N from soil cores, the difference in inorganic N concentrations between the soil at the beginning and end of the incubation is a direct measure of net mineralization or immobilization (Subler et al., 1995). Also, mineralization rates are known to be sensitive to the availability of water (Stanford and Epstein, 1974; Myers et al., 1982; Nadelhoffer et al., 1991). Adams et al. (1989) proposed the use of perforated columns to allow moisture equilibrium across the interface between the bulk soil and the contained soil. The minimal soil disturbance, ease of column insertion and removal from the soil are some of the advantages presented by this technique. This approach also allows the estimate of soil N losses by difference of N content between open and closed columns. Closed columns with a PVC cap on the top prevent water entry during rainfall or irrigation and minimize leaching of mineralized N during incubation.

The soil microbial biomass is an important component of the soil organic matter that regulates the transformation and storage of nutrients (Horwath and Paul, 1994). This is especially true for the N applied in agricultural soils, when immobilization and mineralization processes determine the availability of this nutrient for growing plants, and also may influence the extent of gaseous and leaching losses.

Observation that K_2SO_4-extractable NH_4-N increased after soil fumigation with chloroform ($CHCl_3$) suggested that a procedure developed for evaluation of biomass C could be used to measure biomass N. Assuming that soil fumigation does not affect the mineralization rate of non-biomass organic matter, then the flush of mineral N provides a measure of the amount of biomass N in a soil (Jenkinson and Powlson, 1976; Shen et al., 1984).

Urea is the most common N fertilizer used for various crops around the world. Its large use is due to high content of N, which favors the economics of manufacturing, handling, storage, transportation, and spreading in the field (Havlin et al., 1999). However, N uptake efficiency from urea is quite low if it is not incorporated, because of volatilization of ammonia (NH_3). Gaseous loss from soil following urea fertilizer application may account for up to 75% of total N applied (Fenn and Miyamoto, 1981). On the other hand, NH_3 volatilization from other forms of nitrogen fertilizer is less significant than from urea in non-alkaline soils (Havlin et al., 1999). The general process of N volatilization from applied urea is related to a localized increase in soil pH after dissolution and hydrolysis of the fertilizer (Hauck, 1984).

Marshall and Debell (1980) listed four types of devices used to measure N gaseous losses from soils. Three of these use acid sorbers for trapping ammonia but differ

in the mechanism by which NH_3 and other gases may move from the soil to the sorber: (i) closed-static system (Volk, 1970) which restricts exchange with the atmosphere, thereby creating a closed NH_3 sink; (ii) semi-open system (Nômmik, 1973a) which permits exchange with the atmosphere via diffusion, and (iii) closed-dynamic systems (Watkins et al., 1972) in which NH_3 and other gases are removed from the soil surface by air bubbled through a NH_3 trap. The fourth type involves use of ^{15}N-enriched urea to assess fertilizer N balances, where amounts that cannot be accounted for in known pools are attributed to gaseous loss, presumed to be mainly NH_3 (Nômmik, 1973b). Kissel et al. (1977), Ferguson et al. (1988) and Sherlock et al. (1989) also tested variations of the semi-open system in the field, which allowed continuous measurement of NH_3 volatilization over a broad range of environmental conditions and with a number of different treatments. Estimations were obtained indirectly that up to 47% of applied N may volatilize. In this study, measurements were influenced by environmental conditions such as wind and vapor pressure.

Field evaluation of NH_3 losses can contribute to an accurate estimation of reduction of the efficiency of the applied N fertilizer, which is important for developing an accurate N budget for a given cropping system. The objectives of this research were to (i) evaluate soil net N-mineralization; (ii) examine the relationship between N mineralization and soil microbial biomass N; (i) N leaching losses; and (iv) evaluate NH_3 volatilization from NH_4NO_3 or urea surface applied to a sandy Entisol for citrus trees.

Material and Methods

Characteristics of the Study Area and Experimental Design

The experiment was conducted using 6-yr-old orange trees on a Candler fine sand (hyperthermic, uncoated Typic Quartzipsaments) in Lake Alfred, FL. The Candler soil is a deep well-drained sand with no confining soil horizons, with low organic matter content and low water holding capacity, and consequently is subject to water deficits and needs frequent irrigation. Selected soil physical and chemical characteristics are presented in Table 4.1. 'Hamlin' orange trees (*Citrus sinensis* (L.) Osb.) on Swingle citrumelo rootstock (*Poncirus trifoliata* (L.) Raf. x *Citrus paradisi* Macf.) were planted in September 1993 at 7.6 by 4.6 m spacing (285 trees ha^{-1}).

The experiment had a randomized block design with three replications. Treatments included (i) unfertilized trees, and (ii) trees fertilized with urea or (iii) ammonium nitrate. Single tree plots were used in this experiment. Fertilizer was distributed uniformly as dry granules to the soil surface in a circular area (1.10 m radius) under the tree canopy. Since the scheduled application of dry fertilizer was in four equal doses of the annual amount, 25% of the annual recommended rate (230 g N per tree; Ferguson et al., 1995) was applied on February 15, 1999. Fertilizer application was followed by 7 mm irrigation water using under-the-tree low volume sprinklers to promote fertilizer dissolution and shallow incorporation into the soil. Trees were irrigated using one emitter per tree covering about 7 m^2 area under the tree canopy with a delivery rate of 50 L h^{-1}. During the course of the experiment, irrigation scheduling was programmed when available soil moisture depletion attained 33% in the top 40 cm soil depth based on

Table 4.1. Selected characteristics of Candler fine sand at the experimental site.

Soil characteristics	0-15 cm depth	15-30 cm depth
pH †	7.03	6.98
Organic carbon, g kg^{-1} ‡	6.1	5.1
Total nitrogen, g kg^{-1} ‡	0.4	0.2
P, mg kg^{-1} §	53	42
Bulk density, g cm^{-3}	1.47	1.51
Sand, %	96.7	97.2
Silt, %	0.8	0.5
Clay, %	2.5	2.3

† Water/soil ratio, 2:1 (v/w).
‡ CNS Analyzer, NA-1500, Carlo Erba, Haak-Buchler Instruments, Saddlebrook, NJ.
§ Mehlich 1 extraction.

the continuous measurement of soil moisture using capacitance probes placed at the drip line of citrus trees at various depths (Alva and Fares, 1998; Fares and Alva, 1999). The area under the tree canopy was free of weeds as a result of chemical and manual controls.

Nitrogen Mineralization and Leaching Losses

Soil N mineralization was estimated using an in-situ soil column technique (Raison et al., 1987; DiStefano and Gholz, 1986; Stenger et al., 1996; Dou et al., 1997). Polyvinyl chloride (PVC) columns (35 cm high; 5 cm in diameter) were driven 30 cm into the soil under the tree canopy area, about 80 cm from the irrigation emitter. There were two sets of columns, one closed with a PVC cap on the top to prevent water entry during rainfall or irrigation and to minimize leaching of mineralized N during incubation, while the other column was kept open. The cap was loosely fitted to the PVC column to allow air exchange above the soil with the outside environment. Columns had 8 holes

(diameter = 1 cm) on the walls in order to promote moisture equilibrium between the soil inside the column and the bulk soil outside as described by Dou et al. (1997). Soil moisture content measurements were not statistically different inside and outside the columns.

Columns were driven into the soil after fertilization and every two weeks thereafter during a 75-day period from March 1 to May 11, for a total of five incubation periods. Soil samples also were collected adjacent to the column at 0 to 15 and 15 to 30 cm depths on the day of installation of the incubation columns to estimate the status of available N forms at time zero (initial concentration). At the end of the incubation period, columns were retrieved from the field and divided into 0 to 15 and 15 to 30 cm sections to measure the concentration of both NH_4-N and NO_3-N (final concentration). Ten gram of field-moist soil was weighted into 250 mL centrifuge flasks, 100 mL of 2 mol L^{-1} KCl solution was added. Flasks were agitated on an orbital shaker for 30 min at 2 Hz. The suspension was centrifuged (60 Hz; 10 min) and filtered through Whatman No. 42 filter paper. Extracts were stored in a cold room (4°C) and then analyzed colorimetrically for inorganic NH_4-N (Environmental Protection Agency Method 351.2, 1993) on a Technicon AutoAnalyzer II (Technicon Industrial Systems, Tarryton, NY) and for NO_3-N (Method A303-S170, 1986) on a rapid flow analyzer (ALPKEM Corporation, Clackamas, OR). Nitrogen mineralization at each incubation period was calculated by the increase in the concentration of NH_4-N or NO_3-N in the final as compared to that in the initial sampling. The summation of this amount over successive incubation periods was an estimate of cumulative N mineralization for the total duration.

The difference in the overall N status between the closed and open columns within a given treatment provided an estimated of N leached from the top 30 cm of soil.

The amount of irrigation water applied to the orange trees and the weather conditions (rainfall, air temperature, soil temperature) collected from the FAWN data set (Florida Automatic Weather Network) for the Citrus Research and Education Center in Lake Alfred, FL were recorded from February to May 1999.

Microbial Biomass N

Microbial biomass N (MBN) was determined by the chloroform fumigation-extraction (CFE) technique described by Brookes et al. (1985a, b) in soil samples collected outside the PVC columns starting on March 1. Twenty-five grams of field moist soil samples were transferred to 50 mL glass beakers placed in an amber-glass vacuum desiccator with an additional beaker containing 60 mL of chloroform and several boiling chips. The desiccator was evacuated until 2 min after the chloroform began to boil. This procedure was repeated three times; air was allowed back into the desiccator by means of a screw control valve on the lid after the first two evacuations. After the third evacuation, the desiccator was sealed under vacuum for 24 h. Fumigated and nonfumigated (paired) samples were immediately extracted with 100 mL of 0.5mol L^{-1} K_2SO_4 for 1 h on a orbital shaker (3 Hz) and filtered through Whatman No. 42 filter paper.

Extracts were stored in a refrigerator at 2°C for up to 20 days. Aliquots of K_2SO_4 extracts (25 mL) were subjected to Kjeldhal-N digestion (Bremner, 1996). Recovery of organic N was checked by performing digestions using a solution prepared with primary standard of tris (hydroxymethyl) aminomethane (tradename, THAM®) (Mulvaney, 1992).

Samples were brought to 25 mL with deionized-water. Extracts were analyzed for NH_4-N colorimetrically (EPA Method 351.2, 1993). Microbial biomass N was determined by subtracting the total N extracted in the K_2SO_4 solution of non-fumigated from that of fumigated samples (Brookes et al., 1985a, b). The efficiency of organic microbial N (k_N) for CFE 1 day incubation used to calculate the MBN was 0.54 (Brookes, et al. 1985a).

Ammonia Volatilization

Ammonia volatilized from dry-granular N fertilizers (AN and UR) applied to the soil surface was evaluated using a semi-open static system of ammonia sorbers (Nômmik, 1973a). A modification of the Nômmik's apparatus was made in order to assess NH_3 losses under forced air flux (dynamic system) using a cooling fan (Brusheless Fan, 12 VDC, 7.6 cm diameter; from Radioshack) set inside the trapping device and hooked up to a rheostat. The fan plus rheostat were connected to a 12 V automotive battery. The fan speed was calibrated in the laboratory using a portable anemometer, and manually set to 0.7 m s^{-1}. The trapping device consisted of a PVC-cylinder, 35 cm long and 30 cm in diameter. Two discs of polyurethane plastic foam (density = 0.03 g cm^{-3}; thickness = 2 cm; and circular area = 723 cm^2) soaked in 2.25 mol L^{-1} H_3PO_4 plus glycerol 4.0% (v/v) solution were placed inside the PVC-cylinder at 10 and 20 cm from the top. Each disc retained 120 to 150 mL of the acid-glycerol solution. The lower disc traps the ammonia volatilized from the soil, while top disc serves to trap any atmospheric NH_3, which could contribute to overestimate the NH_3 volatilized from the soil.

Collectors were placed on the soil surface within the fertilized area under the tree canopy immediately after application of the fertilizer and initial irrigation. After fixed periods of exposure, the NH_3 sorber discs were removed and replaced by fresh sorber

discs. The duration of exposure during the first week after fertilization were 9 to 15 h, followed by 2- day period for the next 3 days, and then every 4 or 5-day period for the next 6 days, for a total of 44 days. Discs collected in the field were placed in sealed plastic bags, taken to the laboratory, and stored in a cold room at 4°C. Ammonia trapped by the plastic foam discs at the lower position in the PVC-cylinder was extracted in the laboratory by leaching and squeezing them with portions of 1 mol L^{-1} KCl, and thereafter made up to known volumes (500 or 1000 mL). Extracts were analyzed for NH_4-N colorimetrically (EPA Method 351.2, 1993). Soil temperature was recorded daily at 8 AM and 5 PM within a 10 cm depth layer. Wind speed was also monitored daily using a 3-cup anemometer (Weather Instruments, Princeton, NJ; from Science Associates, Inc.) installed at 0.5 m above the soil level. Soil pH (water:soil ratio = 2:1 v/w) was measured using a glass electrode on samples collected from the 0 to 15 cm soil depth layer during the course of the experiment.

Data Analysis

Data were tested for significant differences among treatments for a randomized complete block ANOVA with three replications using the GLM procedure of the SAS® system (SAS Institute, 1996). Regression analysis was performed with data collected for NH_3 volatilization from fertilized plots using the SAS REG procedure. The model was of the type $y = a + b/x$; where y = N-NH_3 loss (% of applied N), and x = time interval after fertilization (day). Rates of NH_3 evolution during the day and evening were tested using the PROC MIX procedure.

Results and Discussion

Nitrogen Mineralization, Microbial Biomass N and Leaching Losses

Net N mineralization was estimated for incubation periods of about 15 days (Fig. 4.1 and 4.2). An initial net immobilization of N was followed by a period of net mineralization at the 0 to 15 cm soil depth (Fig. 4.1A). Part of the fertilizer N applied to the soils may be converted to organic N as a component of the microbial biomass (Broadbent, 1984). Accumulation of surface decomposed tissue increases the potential for N immobilization since more organic C is available to the soil microbes thus increasing microbial activity. Soil organic-C content was relatively higher at the surface (Table 4.1). Work with ^{15}N reviewed by Legg and Meisinger (1982) suggests that 10 to 40% of N fertilizer may be incorporated in soil organic matter. Figure 4.1B shows increased microbial biomass N for fertilized soil on March 15 as compared to the non-fertilized treatment. On the other hand, fertilization promoted a significant increase in net N mineralization at the 15 to 30 cm soil depth with minor variation on the microbial biomass N following fertilizer application (Fig. 4.2A and B). Nitrogen mineralization was very low at the subsequent evaluation periods at the 15 to 30 cm soil depth. Bauhus (1998) reported for an acid Cambisol covered by a forest floor, values of MBN of about 475 mg N kg^{-1} for an organic horizon and about 30 mg N kg^{-1} for a mineral horizon (5 to 10 cm depth). Regardless of the high acidity presented by the Cambisol, microbial populations could be significant since different species are well adapted to such condition

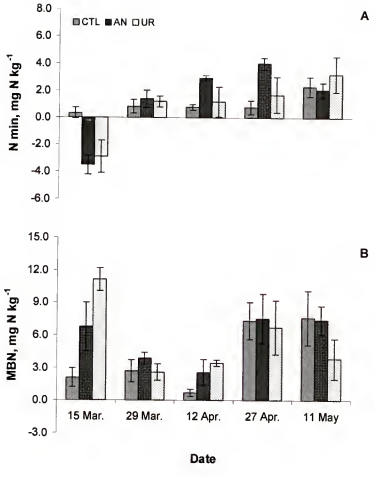

Figure 4.1. Net nitrogen mineralization (Nmin) and microbial biomass N (MBN) in a sandy Florida Entisol under young citrus trees at 0 to 15 cm depth. Fertilizer was applied on February 15. Legend: CTL = unfertilized; UR = urea treated; and AN = ammonium nitrate treated. Vertical bars indicate the standard error of the mean ($n = 3$). a) Nitrogen mineralization vs. time; b) Microbial biomass N vs. time.

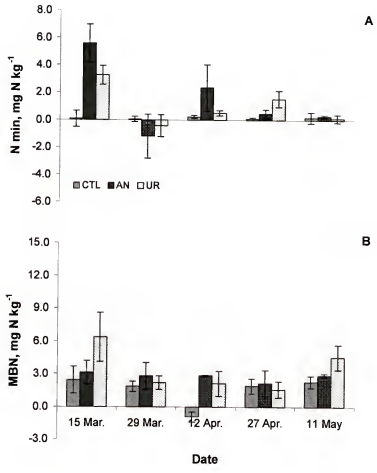

Figure 4.2. Net nitrogen mineralization (Nmin) and microbial biomass N (MBN) in a sandy Florida Entisol under young citrus trees at 15 to 30 cm depth. Fertilizer was applied on February 15. Legend: CTL = unfertilized; UR = urea treated; and AN = ammonium nitrate treated. Vertical bars indicate the standard error of the mean ($n = 3$). a) Nitrogen mineralization vs. time; b) Microbial biomass N vs. time.

associated with an undisturbed environment. Our values for MBN were up to 10 mg N kg^{-1} at the surface soil layer (Fig. 4.1B). Lovell and Hatch (1998) verified that the biomass N, as measured by the chloroform fumigation and extraction method, was unchanged during a short term period after addition of N to a soil with high clay content and mean MBN value of 160 mg N kg^{-1} dry soil. However, the specific respiration, reflecting the metabolic activity of the biomass, showed a decrease immediately after the application of N and was halted following 5 weeks of fertilization, when it was no longer significantly different from the initial value (5 μg CO_2-C mg^{-1} biomass-C g^{-1} h^{-1}). The extent to which inputs of N that can affect microbially mediated processes are not well understood, since additions of fertilizer N could influence soil microbial biomass activity, without its size being measurably affected.

Roots severed after insertion of PVC columns into the soil could influence estimations of net N-min after decomposition of tissue material or by altering the input of root derived carbon (root exudates) and consequently microbial population activity. Raison et al. (1987) assumed that a linear increase of mineral N accumulation in incubated soil indicated that severed roots had no effect on N mineralization estimated using the PVC columns. Bauhus (1998) found no evidence to support the hypothesis that abscission of fine roots could induce N immobilization in a beech forest soil by microbial biomass. In order to minimize interference of severed roots on N-min estimates incubation periods should be kept shorter than 30 or 60 days (Bauhus, 1998). However, if no significant accumulation of inorganic nitrogen is achieved during the incubation period large variation may occur.

Rates of net N-min observed for the non-fertilized treatment were <0.1 mg N per day per kg soil at the 0 to 15 cm soil depth. Our observations are comparable to those reported by Dou et al. (1997) in an experiment carried out with under the canopy of 4-yr-old orange trees on a Tavares fine sand in Florida during February to April 1994. However, the mineralization rates were much greater for older trees, i.e., up to 0.2 mg N per day per kg soil, as reported by Dou et al. (1998). Nitrogen mineralization was greater at the 0 to 15 cm depth compared to that at the 15 to 30 cm depth (Fig. 4.1A and 4.2A). The amount of organic residue deposited on the soil surface by shed leaves, petals, and fruit associated with the greater amount of feeder roots provide conditions for decomposition and turn over of nitrogenous components in the soil.

An increase on N-min was observed for the April 12 and 27 incubation periods as compared to the previous periods (Fig. 4.1A). The N-min on the unfertilized treatment was about 1 mg N kg^{-1} as compared to 1 to 5 mg N kg^{-1} for fertilized treatments during the above-referred interval (Fig. 4.1A). The priming effect of nitrogen fertilizers on plant uptake of soil N has been reported (Broadbent, 1984). Raison et al. (1987) found that, when N mineralization was measured in situ on undisturbed forest soils, N fertilization increased the process 4-fold. However, the priming action can be overemphasized. The major cause of the increased availability of N-min, as measured by ^{15}N plant uptake, is a result of the interchange of fertilizer N for the native humus N (Stevenson and Cole, 1999). After March 15, net N-min occurred at all subsequent incubation periods (Fig. 4.1A). This corresponded to an increase in microbial biomass N over the entire duration of this study (Fig. 4.1B). Daily average soil temperature at 10 cm depth was 21°C during February and March, compared to about 28°C during April and May (Fig. 4.3).

Temperature greatly influences microbial activity and mineralization of soil organic residues, with optimum range for N-min between 25 to 30°C (Stanford et al., 1973). The temperature coefficient, Q_{10}, is 2 over the range 5 to 35°C (Stanford et al., 1977). Thus, a 2-fold change in mineralization rate is associated with a shift of 10°C within this range of temperature. Dou et al. (1997) also found a positive correlation between the quantity of net N-min and daily average temperature on a citrus grove throughout the year.

There was not substantial variation in microbial biomass N at the 15 to 30 cm soil depth during the periods evaluated in the present study (Fig. 4.2B).

The differences in concentrations of NH_4-N and NO_3-N measured between the open and closed columns at each incubation interval represent N losses including denitrification, volatilization, and leaching. Nitrogen losses through denitrification vary greatly and are important mainly under conditions of poor drainage (anaerobiosis) and abundant supply of readily available organic carbon for denitrifying microorganisms (Myrold, 1998; Stevenson and Cole, 1999). The soil used in this study was well drained with irrigation only to replenish the deficit soil moisture, therefore the soil was not likely in anaerobic condition. Thus, denitrification loss was assumed to be insignificant under the conditions of this study. On the other hand, NH_3 volatilization was very important for surface applied urea (as discussed later). Thus, estimation of N leaching losses by mass balance without including an estimation of the volatilization losses would overestimate the leaching losses. Table 4.2 shows the variation of inorganic N forms in the soil during the incubation periods from March 1 to May 11. A great variation of inorganic N was observed during the incubation period ending on March 1, in which 17.3 and 26.5 mg N kg^{-1} were lost at the 0 to 15 cm soil depth for AN and UR treatments, respectively.

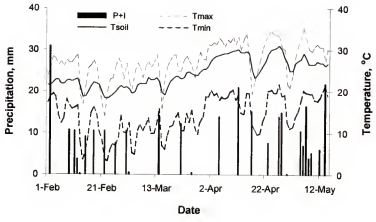

Figure 4.3. Weather data for the experiment site. Legend: P+I = rainfall plus irrigation water; Tmax and Tmin = daily maximum and minimum average air temperature, respectively; Tsoil = average soil temperature (10 cm depth).

Since NH_3 volatilization was greater for the urea treatment, we can infer differences between fertilizer sources were due to gaseous loss rather than leaching losses. An increase in N concentration observed at the 15 to 30 cm soil depth layer for the same incubation period especially for the AN treatment (-6.0 mg N kg^{-1}) as NO_3 leaching was most probable to occur (Table 4.2). Nitrogen from UR fertilizer is not readily nitrified (Havlin, et al. 1999); thus NO_3 leaching was not likely to occur in the first 15 days of observation as compared to that derived from AN (50% NO_3-N). Since NH_3 volatilization was very low after March 15, we may assume that any further variation of N in the soil profile was due to movement with the percolation water. Leaching was effective until March 29 for both soil layers. After this period, the N leaching from the columns decreased substantially (< 1.0 mg N kg^{-1}) (Table 4.2). Total rainfall during April 30 through May 9 was 33 mm. There was a single rain event of 14 mm on March 14 (Fig. 4.3), which could have contributed to greater N losses from the 15 to 30 cm depth soil

Table 4.2. Inorganic N losses at 0 to 15 and 15 to 30 cm soil depth under the canopy of 6-yr-old 'Hamlin' orange tress during incubation using the in situ incubation PVC columns.

Date of Sampling	Incubation	Non-fertilized			AN			UR			LSD ‡
		NH_4-N	NO_3-N	Inorg-N†	NH_4-N	NO_3-N	Inorg-N	NH_4-N	NO_3-N	Inorg-N	
	d	mg kg^{-1}									
						0-15 cm					
1 Mar.	15	0.2	0.6	0.8	9.9	7.4	17.3	24.2	2.3	26.5	6.85
15 Mar.	15	0.2	0.7	0.9	0.7	1.9	2.6	4.4	4.0	8.4	2.67
29 Mar.	15	0.2	0.4	0.6	0.8	1.3	2.1	0.2	1.8	2.0	1.94
12 Apr.	15	0.1	0.4	0.5	0.1	0.5	0.6	0.1	0.4	0.5	1.90
27 Apr.	16	0.1	-0.1§	0	0.1	0.3	0.4	0	0.7	0.7	2.11
11 May	15	0.4	0.4	0.8	0.2	0.5	0.7	0.1	0.3	0.4	1.52
						15-30 cm					
1 Mar.	15	0.2	-0.3	-0.1	0.1	-6.1	-6.0	0.7	-1.6	-0.9	2.11
15 Mar.	15	0.1	0.3	0.4	0.9	6.6	7.5	0	3.1	3.1	2.28
29 Mar.	15	0.2	0.1	0.3	0.6	0.8	1.4	0.2	1.1	1.3	1.54
12 Apr.	15	-0.2	0.1	-0.1	0.4	0.9	1.3	0	0.3	0.3	1.17
27 Apr.	16	0	0.2	0.2	0	0.2	0.2	0.1	-0.3	-0.2	2.07
11 May	15	0.2	0.3	0.5	-0.1	0.3	0.2	0	0.1	0.1	2.11

† Inorg.-N = NH_4-N + NO_3-N.

‡ The least significance difference (P = 0.05) for Inorg-N within fertilization treatments.

§ Negative numbers indicate accumulation of N.

recorded for the March 15 (7.6 and 3.1 mg N kg^{-1} for AN and UR, respectively). The irrigation system used in the experimental site was set to replenish the soil moisture content within the rooting depth when moisture content fell below recommended levels, and did not contribute to further leaching of nitrate below the 30 cm soil layer.

Ammonia Volatilization

Data of cumulative NH_3 losses from applied AN or UR fertilizers during 44 days of field evaluation are presented in Fig. 4.4. The response model fitted to measure values for each of the fertilizer source showed R^2 values of 0.76 to 0.78 ($P<0.001$). Volatilization losses were much greater for UR as compared to that for AN. Volatilization of ammonia increased rapidly during the initial 5 days after N source application. However, in the case of UR, volatilization continued at low rates until 10 days after application.

Differences of NH_3 volatilization from AN and UR source was also related to differences in soil pH. Average soil pH with AN application was 6.9 ± 0.1, which was similar to that of unfertilized soil. In the case of UR amended soil, the pH initially increased to 8.4 ± 0.4, and took about 32 days to return to the original pH. Volatilization of NH_3 depends on the quantity and equilibrium of NH_3 and NH_4 forms in the soil solution, which are highly dependent on soil pH. The relative concentration of NH_3 (pKa = 9.3) increases 10% and 50% at pH 8 and 9.3, respectively (Havlin et al., 1999). For AN, the overall soil pH is important to determine NH_3 volatilization losses whereas the hydrolysis of urea causes a marked increase in the soil pH close to the fertilizer. The soil pH in the immediate vicinity of the urea granules may reach values of up to 9 (Havlin et al., 1999), what shows the importance of chemical reactions of N in soil microsites

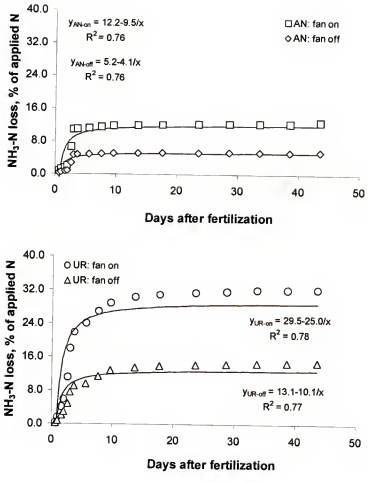

Figure 4.4. Ammonia losses as measured by a semi-open collector system from field fertilized with ammonium nitrate (AN) or urea (UR). Ammonia collector system set with (fan on) and without (fan off) additional air circulation.

(Hauck, 1984). Ammonia volatilization is favored in sandy soils with low buffering capacity, since the ability of NH_4 to form electrostatic bonds with clay minerals and organic colloids is low to impair losses of soil and fertilizer N (Stevenson and Cole, 1999).

The NH_3 volatilization losses from both N sources were greater with additional air inside the collector to simulate the ambient air movement as compared to that with no additional air circulation. About 5.5 and 12.8% of N applied as AN (16 kg N ha^{-1}) were lost by NH_3 volatilization without or with additional air circulation inside the collectors, respectively. The corresponding data for urea were 14.9 and 32.3%. Average wind speed measured on the citrus grove was 0.7 m s^{-1} (range of 0.4 to 1.3 m s^{-1}) during the initial 7 days of fertilizer application. A similar condition to the initial week occurred from 7 to 44 days after fertilization, when average wind speed was 0.6 m s^{-1} (range of 0.4 to 0.9 m s^{-1}). Environmental conditions (aeration, temperature, and soil moisture) can have marked influence on the measurements made in the field. Wind speed controls NH_3 volatilization through its effect on the rate of transport of NH_3 away from the soil-air interface (Peoples et al., 1995). Ammonia volatilization is driven by the difference in NH_3 partial pressure between the atmosphere and that in equilibrium with the moist soil. Marshall and Debell (1980) reported that lowest NH_3 losses from UR fertilizer were estimated in field experiments by a close-static (13%) and a semi-open (17%) methods, which restricted airflow as compared to a closed-dynamic method (22-26%). Artificial conditions created in a close environment can hinder NH_3 diffusion from the soil surface to the sorbers and may cause NH_3 re-adsorption by the soil.

The NH_3 evolution rates during the first week after N fertilizer application to the soil surface is shown in Table 4.3. Reported averages were significantly different for the day and evening periods and lower than NH_3 peak flux (40 to 110 mg NH_3-N m^{-2} h^{-1}) measured following urea application to a ryegrass/white clover pasture as observed by Black et al. (1985a). Such patterns are closely related to soil temperature differences since deviations for NH_3 losses were observed for measurements made by the semi-open system using the fan-off or the fan-on, irrespective to the period evaluated. Soil temperature measured in the 10 cm depth layer was 18.5 ± 3.3°C at 5 PM, and 14.3 ± 2.4°C at 8 AM, which represents a difference greater than 4°C. Ferguson et al. (1988) reported that maximum rates of NH_3 loss from UR solution applied to the soil were observed near midday, when water content at the soil surface was beginning to decline and the surface temperature was rapidly rising. Similarly, Lightner et al. (1990) reported that the peak of urea-N losses through NH_3 volatilization occurred between midmorning and early afternoon. This corresponded to periods of increasing air temperature and moisture flux toward the soil surface. Volatilization increases with increasing temperature, which is related to higher reaction rates and urease activity. Voss (1984) reported that the activity of such enzyme is enhanced up to the limit of 70°C.

Lara-Cabezas et al. (1999) estimated the collector efficiency factor (E) for a semi-open static system similar to that used by Nômmik as 1 to 50%, by estimating the ratio of volatilized NH_3 retained in the collector with ^{15}N mass balance method. The authors found models that would correct the lower efficiency of the semi-open system. Then, estimated losses in the present study could be lower than the real condition, especially for the system that had no fan installed in the collector.

Table 4.3. Ammonia volatilization rates from either ammonium nitrate (AN) or urea (UR) from a Candler fine sand during a week after fertilizer application.

N source	Fan condition	Day †	Evening †	P<F
		mg NH_3-N m^{-2} h^{-1}		
AN	Off	5.2	2.7	0.1208
	On	12.0	6.0	0.0062
UR	Off	9.4	5.3	0.0044
	On	22.9	12.7	0.0407

† Mean value (n = 4).

Conclusion

Nitrogen fertilization affected slightly soil N mineralization/immobilization processes as compared to the non-fertilized treatment, and could interfere with estimates of soil available N for tree growth and fruit yield. Soil N immobilization was observed only during the incubation period that started 15 days after fertilization. Microbial biomass N (0-15 cm depth) increased by N fertilization during the 30 days after fertilizer addition. Leaching losses of N from the 15 to 30 cm soil depth layer were < 9 mg N kg^{-1} using a continuous monitoring of soil moisture and irrigation system. The PVC column technique used for N leaching estimates was influenced by NH_3 losses of added N from the soil. Ammonia loss was greater from UR as compared to that from AN. Volatilization accounted for 5.5 and 12.8% of applied N as AN or UR, respectively, without additional air circulation inside the collection chamber. The losses increased to 33.3% with additional air circulation inside the sample chamber.

CHAPTER 5
BIOMASS DISTRIBUTION AND ^{15}N PARTITIONING IN CITRUS TREES ON A SANDY ENTISOL

Introduction

Best management practices (BMP) for citrus have been proposed in order to improve the efficiency of nitrogen (N) utilization for high fruit yield and also to address environmental quality issues in regard to N contamination of the groundwater in Florida. Efficient management of nitrogen fertilization and irrigation represent two important cultural factors responsible for optimal production.

Perennial trees store large quantities of N within various plant components, which can be utilized for tree growth and fruit yield in subsequent seasons, as reported for almond trees (Weinbaum et al., 1984), citrus (Legaz et al., 1995), kiwifruit (Ledgard and Smith, 1992), and apple (Millard and Neilsen, 1989; Khemira et al., 1998). The annual growth and the fruit yield of citrus trees contain a small proportion of the fertilizer N applied during the growth period. The greatest amount of N in the new growth is drawn from the tree biomass, therefore, the N reserve in the older tissues plays an important role in the development of new leaves and flowers in the spring, when uptake from the soil is low (Kato, 1986). Despite the fact that N on the tree skeleton is considered as a major N pool in the soil-plant system there is no clear definition of nitrogen storage within tree components.

Dry matter and N distribution in mature citrus trees vary with tree age and variety. Legaz et al. (1981) reported that 'Valencia' orange trees grown outdoors in sand culture, which were fertilized with a nutrient solution labeled with ^{15}N-nitrogen, presented distribution of N absorbed among tree parts ranked in the order (mg ^{15}N per 100 mg ^{15}N in the whole tree): leaves (49.7) > roots (19.2) > twigs and stem (14.3) > flowers (10.6) > ovaries (6.1). Moreno and Garcia-Martínez (1984) described the biochemical process of N mobilization in citrus trees, in which total protein content of old leaves decreased progressively from the beginning of February (spring flush) to June. Almost 90% of the mobilized protein came from an aqueous protein that was the major fraction (50-70%) of the total leaf protein in Washington navel sweet orange.

The removal of N by plant biomass has been used to estimate N requirements, mainly for annual grain crops. In contrast, the large pool of N present in the structural framework of citrus trees implies that distribution and remobilization of N within the tree play an important role in determining the N requirement on annual basis to maximize N uptake efficiency and minimize its losses (Sanchez et al., 1995).

Efficient management of irrigation can minimize leaching losses of highly soluble nutrients (i.e., nitrate) through the soil profile below the rooting zone. Citrus grown in deep sandy soils with high volume irrigation systems, as used in central Florida in the past, tends to dry the upper layers between irrigation at long intervals. This condition favors deep rooting as reported by Cahon et al. (1962), Castle and Krezdorn (1977), Castle et al. (1993), and Boman et al. (1999). The same occurs in nonirrigated groves, where the majority of the rooting system can reach depths >1.5 m in the soil (Pace and Araujo, 1986; Oliveira et al., 1998). However, using low volume irrigation systems to

replenish the deficit moisture in the surface soil may lead to a shallow rootzone of the citrus trees. Since citrus growth and distribution can be modified as a result of changes in the root environment, a clear understanding of the root system is important to develop irrigation and nutrient BMPs in an effort to improve uptake efficiency and minimize losses below the rooting depth.

The objectives of this study were (i) to evaluate biomass distribution of 6-yr-old citrus trees grown in a sandy soil under low volume irrigation, and (ii) to estimate partitioning of soil applied-^{15}N during early spring in different plant components during fruit harvest.

Material and Methods

Site Characteristics and Treatments

A citrus grove with 'Hamlin' orange trees (*Citrus sinensis* (L.) Osb.) on Swingle citrumelo rootstock (*Poncirus trifoliata* (L.) Raf. x *Citrus paradisi* Macf.) planted in September 1993 at 7.6 by 4.6 m spacing (285 trees ha^{-1}) on a Candler fine sand (hyperthermic, uncoated Typic Quartzipsamnets; sand = 96.7%; silt = 0.8%; clay = 2.5% on the top 15 cm layer) in Polk County, FL, was used in this study. Selected chemical characteristics are presented in Table 5.1. The experiment was initiated in February 1999 and continued until December 1999, and it was a randomized block design with three replications. Three uniform trees were assigned to each experimental plot.

Fertilizer treatments included: (i) urea-^{15}N, and (ii) $^{15}NH_4{}^{15}NO_3$. The labeled fertilizers presented an isotopic enrichment of 10 atom%^{15}N. Fertilizers were uniformly

Table 5.1. Selected soil chemical characteristics at the 0 to 15 and 15 to 30 cm soil depths.

Soil depth	C †	Total N†	pH ‡	P §	K §	Ca §	Mg §
cm	-------- g kg^{-1} --------			mg kg^{-1}	---------- $cmol_c$ kg^{-1} ----------		
0-15	6.1	0.4	7.0	53	0.02	1.7	0.3
15-30	5.1	0.2	6.8	42	0.01	0.8	0.2

† CNS Analyzer, NA-1500, Carlo Erba, Haak-Buchler Instruments, Saddlebrook, NJ.
‡ Water/soil, 2:1 ratio (v/w).
§ Mehlich 1 extraction.

distributed as dry granules to the soil surface in a circular area (1.10 m radius) under the tree canopy in the amount of 25% of the annual recommended rate of 230 g N per tree (Fergunson et al., 1995) on February 15. Following fertilizer application, the area received 7 mm of irrigation water using under-the-tree low volume sprinklers to promote fertilizer dissolution and shallow incorporation into the soil. Trees were irrigated using one emitter per tree covering about 7 m^2 with a delivery rate of 50 L h^{-1}. During the course of the experiment, irrigation was initiated based on 33% depletion of the available soil moisture within 40 cm soil depth determined by a multisensor capacitance probe placed at the drip line of citrus trees (Alva and Fares, 1998; Fares and Alva, 1999).

Two other applications of ^{14}N were made on June 8 and September 9, 1999 (in equal amounts of 86 g N per tree) in order to supply the trees with the remaining annual N recommended rate. Phosphorus and potassium were applied at 40 g per tree P_2O_5 and 120 g K_2O per tree, respectively, also in June and September.

Tree Sampling and Biomass Estimation

Tree flowering started late February 1999 and the spring flush showed complete expanded leaves in April 1999. Leaves and fruit were collected in the experiment following fertilizer application in February 1999. The mature flush of leaves (>8-month-old) and the summer/fall 1998 (3 to 8-month-old) leaf flush were sampled on the day of fertilization (day 0), 7, 14, 21, 28, 35, 49, 63, and 77 days after fertilization (DAF). The summer/fall 1998 component was still sampled at 113 and 206 DAF. Flush of spring 1999 (<3-month-old) was sampled at 49, 63, 77, 113, and 206 DAF. Fruit samples were taken in April (63 DAF; 20 per tree), June (113 DAF; 10 per tree), and September 1999 (206 DAF; 10 per tree), when they showed the average diameter of 1.5, 3.5, and 5.0 cm, respectively. Leaf samples on each sampling date comprised 10 leaves per tree. Leaves and fruit were washed in detergent solution and thoroughly rinsed in tap water and distilled water, and then dried at 65°C for 72 h (fruit were sliced in small pieces before drying). The dried tissue material was ground to pass a 40 mesh screen using a ball mill. To reduce the likelihood of cross-contamination, all samples were ground in order from estimated least ^{15}N concentration to greatest ^{15}N concentration.

The concentration of N and the N isotopic ratio of tissue material were determined with a C/N analyzer linked to a Tracer Mass Isotope Ratio Mass Spectrometer (Europa Scientific) and an automated ^{15}N Analyzer (ANCA-NT) at the Stable Isotope Research Unit of the Oregon State University. The tracer mass system is capable of analyzing with a precision of 0.8 atom ‰ ^{15}N for levels above the natural abundance.

The trees that received the ammonium nitrate fertilizer were destructively harvested in December 1999 for dry matter distribution evaluation in different tree components and sampled for determinations of N concentration and isotopic ratio as described above. The aboveground (AG) portion was divided in (i) summer/fall 1999 leaf flush (<6-month-old); (ii) spring 1999 + older leaves; the latter component was most made up by summer/fall 1998 flushes; (iii) twigs >1.5 cm in diameter; (iv) twigs ≤1.5 cm in diameter; (v) trunk; and (vi) fruit. Soil were excavated in two opposing quadrants (NW and SE) of 1.75 x 1.75 m each, marked on the soil surface, and which had the tree trunk in a common vertex. Then, roots removed from the depths of 0 to15, 15 to 30, and 30 to 45 cm were separated with a 0.2 cm sieve the following size classes: (i) fibrous roots (<0.2 cm diameter), (ii) woody roots (0.2 to1.0 cm diameter), and (iii) woody roots (>1.0 cm diameter). The taproot was also separated from the soil and together with the roots, it comprised the belowground (BG) portion of the tree biomass.

Samples from tree components were collected in the field and placed in sealed plastic bags to prevent water loss, then weighted in the field. Later the same material was washed in the laboratory and dried in an oven (65°C; 72 h) for dry weight determination, and further N analysis as described earlier. Total dry weight of roots was estimated by multiplying the values obtained for both excavated soil quadrants by 2. Fruit subsamples were also sent to the University of Florida, Department of Citrus Packing House for analysis of juice quality (total soluble solids, citric acid content, and soluble solids/acid ratio) according to standard procedures (Wardowski, 1990).

Trees that received the urea (UR) fertilizer had the aboveground portion destructively harvested. However, only the total fresh weight of leaves plus twigs was

obtained. The fresh weight of individual tree components was assumed to be proportional to the corresponding components of those trees treated with ammonium nitrate (AN). Fruit and trunk biomass were individually evaluated. Then, dry matter of components was calculated based on the moisture content of samples collected in the field and dried in an oven (65°C; 72 h). The root excavation was not done from this treatment, since we assumed similar magnitude of root distribution for these trees as that for the trees treated with ammonium nitrate. Nitrogen concentration and N isotopic ratios were measured in each of tree parts.

The percentage of N in the plant components derived from the labeled fertilizer (eq.1) and the total amount of N recovered in different plant components (eq.2) were calculated using the isotopic dilution equations described by Hauck and Bremner (1976):

$$\%N_{dff} = (a-c)/(b-c)*100 \quad [1]$$

where a = atom^{15}N% abundance in fertilized plant material, b = atom%^{15}N abundance of labeled N fertilizer, and c = atom%^{15}N abundance of non-fertilized plant material (background concentration).

$$\% \text{ fertilizer N recovery} = (a-c)/(b-c)*N_p/N_f*100 \quad [2]$$

where N_p = total amount of N in the tree components in grams; and N_f = total amount of ^{15}N applied as labeled fertilizer in grams.

Root Distribution

Roots were also sampled in December 1999 using a 5-cm diameter PVC corer for root density and average root diameter estimations. Samples were collected at 0 to 15, 15 to 30, and 30 to 45 cm soil depths every 50, 100, and 150 cm distance from the tree trunk in the N-S (within row), and E-W (between rows) directions. Samples were taken to the laboratory in sealed plastic bags and separated with 0.2 cm sieve. Two classes of root size were separated using a forceps: (i) <0.2 cm in diameter and (ii) 0.2 to 1.0 cm in diameter. The roots were cleaned and fresh weight was recorded. Root length was determined by counting the number of horizontal and vertical intersections of roots in a grid system of 1.0 x 1.0 cm (Tennant, 1975), which multiplied by 11/14 and divided by the volume of the PVC corer (295 cm^3) gives the root density in cm cm^{-3} soil. Mean root radius was calculated assuming that fresh roots have a density of 1 Mg m^{-3} by the equation $r_0 = (F_{wr}/\pi L)^{1/2}$, where F_{wr} is the fresh weight of roots (g), and L is the total root length (cm) (Barber, 1995).

Data analysis

Standard deviations were calculated for mean N_{dff}, dry matter distribution of tree components, and average root density for each soil depth and trunk distance. A simple analysis of variance (ANOVA) was used to test the hypothesis that means from aboveground biomass distribution of fertilized trees, root distribution obtained for soil quadrants, N content, ^{15}N enrichment, and ^{15}N recovery of tree components are equal ($P = 0.05$) using the GLM procedure of the SAS® system (SAS Institute, 1996).

Results and Discussion

^{15}N nitrogen Uptake After Fertilizer Application

Early effects of ^{15}N application were detected by evaluating percentages of nitrogen derived from the fertilizer (N_{dff}) in the leaves (Fig. 5.1A and 5.2A). Maximum values of N_{dff} observed were about 40% (Fig. 5.1A), indicating the importance of other N sources (tree reserve and soil) for the citrus trees. Such limited contribution of fertilizer-N was also observed by Sanchez et al. (1992) for established pear trees. A larger proportion of N from either the ammonium nitrate or urea labeled fertilizers occurred in younger leaves (up to 38-22%), especially for the spring 1999 flush, compared to mature leaves (up to 12-7%) (Fig. 5.1A and 5.2A). The N remobilization process involves several biochemical steps of protein degradation and translocation into different tree components (Titus and Kang, 1982; Kato, 1986; Engels and Marschner, 1995). Such mobilized N may not be enough to support strong N sinks, and then newly absorbed soil N appeared in higher proportion in new growing tissues. Kato et al. (1982) showed that in the coldest season, the N uptake by satsuma mandarin trees was about one tenth of the summer amount. More than 90% of the applied N was found in the roots in the winter; on the other hand, in the summer, 55% of the absorbed N translocated upwards and most of it was found in the developing new shoots. This suggests that the N taken up was translocated to the aboveground portion of the tree due to the high demand for N in protein synthesis of new developing organs as discussed by Legaz and Primo-Millo (1984) and Kato (1986). Maximum root absorption efficiency is reported to occur in late spring and early summer for peach trees (Muñhoz et al., 1993). The $\%N_{dff}$ increased gradually until March 15 for UR and May 5 for AN fertilized trees. The observed

difference could be due to the ammonia losses from applied UR fertilizer, and consequently a lower availability of the nutrient for plant uptake. Volatilization accounted for 14.9 and 32.3% of applied N for ammonium nitrate and urea, respectively (Chapter 4). Since we assumed that tree biomass distribution was similar for UR and AN fertilized trees (see discussion on tree biomass partitioning), the differences in $\%N_{dff}$ between treatments could be due to differences in the amount of N absorbed after fertilization. A plateau for N_{dff} was reached after May 4, and the further decrease was probably associated with the uptake of nonlabeled N applied on June 8 and September 9 (Figure 5.1A and 5.2A).

The total N concentration in the leaves of the orange trees prior to fertilization was about 21 g N kg^{-1} (Figure 5.1B and 5.2B), and increased for 4 weeks after application of the labeled fertilizer when levels of 26 g N kg^{-1} for the summer/fall 1998 flush for both treatments were observed (Fig. 5.1B and 5.2B). Then, the N concentration declined after March 15 probably as a result of combined processes of N redistribution from mature tissue and leaf expansion of young tissue. Nitrogen redistribution was evident since the concentration in mature leaves of AN treated trees (25.3 g N kg^{-1}) was higher than that of UR treated ones (21.7 g N kg^{-1}), while no major difference appeared for the summer/fall 1998 leaf flush as presented above. By March 15 the residual soil inorganic ^{15}N was very low (data not shown) for significant uptake and maintenance of the N_{dff} proportions in the leaves.

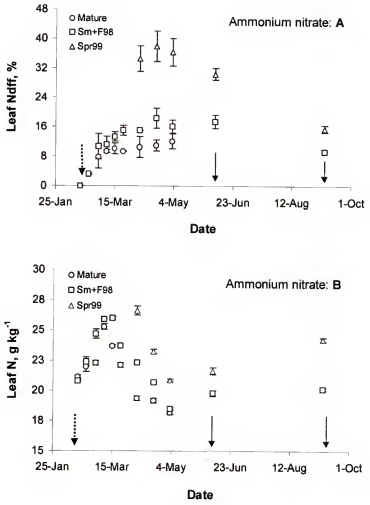

Figure 5.1. Nitrogen in leaf samples derived from the labeled ammonium nitrate fertilizer after application. Vertical bars are the standard error of the mean ($n = 3$). Legend: Mature = oldest leaf flush (>8-month-old); Sm+F98 = summer/fall 1998 leaf flush (3 to 8-month-old); and Spr99 = spring 1999 leaf flush (<3-month-old). Dashed Arrow indicates time of ^{15}N application. Other arrows indicate further application of nonlabeled fertilizer. a) Percentage of N derived from the labeled fertilizer; b) Leaf N concentration.

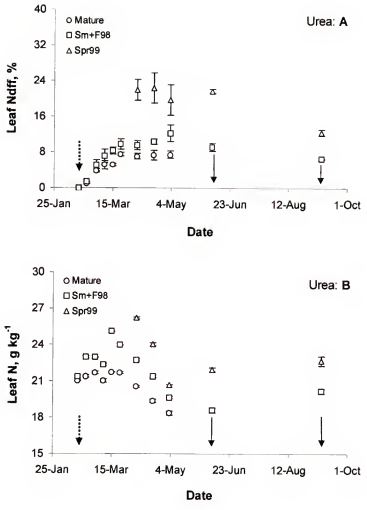

Figure 5.2. Nitrogen in leaf samples derived from the labeled urea fertilizer after application. Vertical bars are the standard error of the mean (n = 3). Lake Alfred, 1999. Legend: Mature = oldest leaf flush (>8-month-old); SM+F98 = summer/fall 1998 leaf flush (3 to 8-month-old); and Spr99 = spring 1999 leaf flush (<3-month-old). Dashed arrow indicates time of ^{15}N application. Other arrows indicate further application of nonlabeled fertilizer. a) Percentage of N derived from the labeled fertilizer; b) Leaf N concentration.

Tree Biomass Distribution

Fertilized trees (n = 6) were destructively harvested when fruit reached maturity. The juice analysis of fruit samples taken in December 1999 showed total soluble solids = 10.5 ± 0.07, acidity = 0.69 ± 0.04% and soluble solids/acid ratio = 15.3 ± 1.14. The mean height of the trees was 2.5 ± 0.01 m with a canopy diameter of 2.5 ± 0.14 m. Trunk height was 41.4 ± 3.9 cm, and perimeter was 24.5 ± 1.5 cm, above the bud union. The aboveground (AG) dry weight estimated for ammonium nitrate or urea fertilizer treatments was >70% of the total tree biomass (Table 5.2 and 5.3). The largest proportion of total tree biomass was that of fruit, which represented 28 to 30%. The average fruit yields were 15,040 ± 1,145 kg ha^{-1} for the ammonium nitrate and 14,750 ± 1,145 kg ha^{-1} for the urea treatments (fresh weight basis). The N source effect was nonsignificant on the fruit yield. Since trees were uniformly fertilized before labeled fertilizer application, it is often difficult to verify growth differences in a single year (Sanchez et al., 1995). Leaves accounted for about 13% of the AG biomass. The summer + fall 1999 flush accounted for a major portion of total leaf dry weight (Table 5. 2). Stansly et al. (1996) reported the seasonal flushing pattern of 4-yr-old grapefruit trees, in which approximately 20% of leaf area at the end of a growing season was carryover from the previous year and only 25% came from spring flush compared to 33% in summer and 35% in fall. This evidence is in conformity with our observation for the greatest proportion of biomass of the summer plus fall flushes in December 1999. The prevalence for summer and fall flushes can be expected with a larger size of individual leaves as compared to mature or spring flush leaves.

Roots accounted for 27.7% of the total weight of the tree. Fibrous roots accounted for the greatest proportion in the belowground (BG) portion (35.6%) after the taproot (38.4%) (Table 5.2). There was a high concentration of fibrous roots in the upper layer of the soil profile. Over 70% of fibrous roots were found in the 0 to 15 cm depth, and at the subsurface depths the root distribution data showed substantial variation as evident from greater coefficients of variation (Table 5.2). Woody roots followed a similar pattern as described above, and represented less than 26% of the total root system (as in the BG portion) within the 0 to 45 cm soil depth layer. Dasberg (1987) reported that the aboveground portion of 9 to 20-yr-old citrus trees including leaves, fruit, trunk, and branches accounted over 65% of the tree biomass. The dry matter partitioning of 22-yr-old Shamouti orange trees (Feigenbaum et al., 1987) was: branches and twigs, 30.1%; trunk plus small branches, 25.3%; roots, 24.0%; fruit, 13.3%; and leaves, 7.3%. In the case of nonbearing (32-month-old) 'Hamlin' trees roots, trunk, large branches, leaves, and small branches accounted for 28.1, 26.1, 21.2, 18.0, and 7.8%, respectively of the tree biomass (Alva et al., 1999). Our data are comparable to the above values since >70% of the total tree biomass was found on the aboveground portion. Proportions of the dry matter of trunk or leaves deviate from reported values by Feigenbaum et al. (1987) and Alva et al. (1999) since the former presented a total value for trunk and main branches, while the latter harvested trees with no fruit. Swingle citrumelo is a superior rootstock for sweet oranges that produce high fruit yields under irrigation (Wutscher and Bistline, 1988; Castle et al. 1993). Such high yields, when related to an induced smaller canopy volume of trees, can explain the greater proportion of fruit on tree biomass as compared to the value reported by Feigenbaum et al. (1987).

Table 5.2. Mean biomass distribution of 6-yr-old 'Hamlin' orange tree on Swingle citrumelo rootstock fertilized with ammonium nitrate.

Tree component	DM †	sd ‡	AG §	BG ¶	Total #
	-------- g --------		---------- % ----------		
Summer + fall 1999 leaf flush	1567	211	8.7	-	6.3
Spring 1999 + older leaf flush	847	116	4.7	-	3.4
Twigs >1.5 cm diameter	2267	317	12.5	-	9.1
Twigs ≤1.5 cm diameter	4249	760	23.5	-	17.0
Trunk	1566	105	8.7	-	6.3
Fruit (9-month-old)	7580	202	41.9	-	30.3
0-15 cm soil depth					
Fibrous roots (≤0.2 cm diameter)	1771	66	-	25.5	7.1
Woody roots (0.2-1.0 cm diameter)	565	55	-	8.1	2.3
Woody roots (≥1.0 cm diameter)	461	314	-	6.6	1.8
15-30 cm soil depth					
Fibrous roots (≤0.2 cm diameter)	611	219	-	8.8	2.4
Woody roots (0.2-1.0 cm diameter)	413	188	-	5.9	1.6
Woody roots (≥1.0 cm diameter)	271	137	-	3.9	1.1
30-45 cm soil depth					
Fibrous roots (≤0.2 cm diameter)	87	55	-	1.3	0.3
Woody roots (0.2-1.0 cm diameter)	50	28	-	0.7	0.2
Woody roots (≥1.0 cm diameter)	55	50	-	0.8	0.2
Taproot	2667	827	-	38.4	10.7
Total AG	18076	349	100.0	-	-
Total BG	6952	580	-	100.0	-
Grand Total	25028	482	-	-	100.0

† Dry matter of tree component.
‡ The standard error of the mean (n = 3).
§ Proportion of the aboveground dry matter.
¶ Proportion of the belowground dry matter.
# Proportion of the total dry matter.

Table 5.3. Mean biomass distribution of 6-yr-old 'Hamlin' orange tree on Swingle citrumelo rootstock fertilized with urea.

Tree component	DM †	sd ‡	AG §	Total ¶
	--------- g --------		-------- % -------	
Summer + fall 1999 leaf flush	1660	114	8.8	6.5
Spring 1999 + older leaf flush	896	61	4.8	3.5
Twigs >1.5 cm diameter	2396	164	12.8	9.3
Twigs ≤1.5 cm diameter	4481	307	23.9	17.4
Trunk	2037	468	10.9	7.9
Fruit (9-month-old)	7297	596	38.9	28.4

† Dry matter of tree component.
‡ The standard error of the mean (n = 3).
§ Proportion of the aboveground dry matter.
¶ Proportion of the total dry matter, assuming the biomass of roots is similar to that of the ammonium nitrate treatment.

Root Density Distribution

Trees showed uniform fresh root length density (L_v, cm root cm^{-3} soil) in all four directions evaluated ($P>0.05$). Root density was greater closer to the tree trunk on both horizontal and vertical planes ($P<0.05$) (Fig. 5.3A and B). Root density decreased from 1.85 at 0 to15 cm to 0.16 cm cm^{-3} at 30 to 45 cm depth within 50 cm from the tree trunk. At 150 cm from the trunk, the root density was lower by 48% as compared to that at the 50 cm distance from the trunk in the 15 cm depth soil (Fig. 5.3A). The same pattern was found for each soil layer evaluated. Barber (1995) listed values for L_v for different annual growing species (i.e., corn, soybean, wheat, oat, barley, and rice), which ranged from 2 to 5 cm cm^{-3} at the upper 15 cm soil depth, while the figure reported for cotton was 1.5 cm cm^{-3}. Root length density of apple trees varies considerably in the top 1.0 m of

soil, and ranges from zero to about 1.0 cm cm^{-3} (Hughes and Gandar, 1993; De Silva et al., 1999). Our values of L_v approach the maximum root length density of 1.6 cm cm^{-3} obtained in the surface 0-15 cm layer for cotton 99 days after planting (Schwab et al., 2000).

Fibrous roots, with mean diameter of 0.08 ± 0.01 cm accounted for a major portion of the root density. Root density for large roots (0.2 to 1.0 cm) showed high variability as shown by the high standard deviation of means in Fig. 5.3B. The root distribution presented for these trees is characteristic for low volume irrigated trees as affected by water and nutrient availability (Spiegel-Roy and Goldschmidt, 1996). In a sandy soil, using continuous monitored low volume irrigation with emitters positioned under the tree canopy, roots will tend to limit their growth to the soil wetting volume. Zhang et al. (1996, 1998) found that root density (dry weight basis) was significantly greater near the emitter and at the 0 to15 cm depth layer for grapefruit trees. Data presented points out the dynamic character of citrus root system and its apparent adaptation to soil texture and irrigation as discussed by Alva and Tucker (1997).

Tree ^{15}N Recovery in December 1999

Nitrogen concentration was lowest in the trunk and taproot (Table 5.4). The N concentration of twigs (4.0 to 7.8 g N kg^{-1}) and roots (5.8 to 17.0 g N kg^{-1}) varied depending on the tissue age. Younger roots had greater N concentration as compared to older roots. Nitrogen concentration in the fruit showed least variation with values around 8.3 g N kg^{-1}, while that of the leaves varied from 21.0 to 25.5 g N kg^{-1}. The values presented above are in agreement with those reported by Chapman (1968).

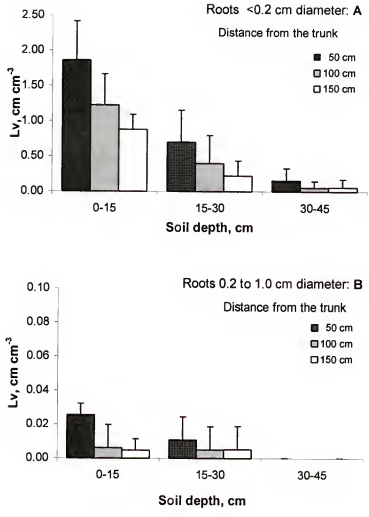

Figure 5.3. Root length density (L_v) distribution of 6-yr-old 'Hamlin' orange trees on Swingle citrumelo rootstock in a sandy Entisol. Vertical bars represent the standard error of the mean ($n = 12$). a) Class of roots < 0.2 cm in diameter; b) Class of roots between 0.2 and 1.0 cm in diameter.

The highest enrichment of labeled N (atom%^{15}N) was found in the fruit, i.e., 1.97 and 1.26 for AN and UR, respectively (Table 5.5). The percentage enrichment of labeled N in the vegetative portion of the trees decreased in the order: leaves > twigs > roots > trunk > taproot (Table 5.5). Akao et al. (1978), Legaz et al. (1982), Feigenbaum et al. (1987), and Legaz and Primo-Millo (1988) reported similar trends for citrus trees.

Nitrogen recovery from the labeled N source was greater for ammonium nitrate (39.5%) as compared to that for urea (25.5%) (Table 5.6). Recovery may have been slightly underestimated since roots were not totally collected from soil, and there was also some loss of N due to senescence and shedding of mature leaves, petals, and young fruit. Feingenbaum et al. (1987) reported the ^{15}N-uptake efficiency from labeled KNO_3 source for 22-yr-old Shamouti orange trees as 40%. In their study, labeled fertilizer was applied with irrigation water in five monthly applications from April to August. Boaretto et al. (1999) found 33% of ^{15}N recovery from urea applied to the soil for 1-yr-old 'Pera' orange trees cultivated in closed pots where leaching losses of nitrate was avoided.

Most of the total ^{15}N recovery occurred in the aboveground portion of the tree and contributed 32.8 and 18.9% of applied N for AN and UR treatments, respectively (Table 5.6). In contrast, Legaz et al. (1982) reported that 50 to 60% of total tree ^{15}N recovery in the aboveground portion of 5-yr-old calamondin trees (*Citrus mitis* Bl.) grown in pots filled with siliceous sand and irrigated with a modified Arnon and Hogland nutrient solution. Although the total N concentration of fruit was similar between ammonium nitrate and urea treatments, the values of N_{dff} were significantly different ($P<0.01$). Plants treated with ammonium nitrate showed N_{dff} values of 35.2, 25.7, 19.8 and 16.6% from April to December, whereas for the urea treatment, the respective values were 18.5, 14.6,

9.3 and 9.2% (Fig. 5.4A). This suggests the fruit relied on N from reserve organs at least during early growth. Fruit was responsible for the highest recovery of applied ^{15}N, followed by leaves, twigs, and trunk and then root biomass (Table 5.6). Experiments with ^{15}N labeled fertilizers showed that the highest rate of N uptake by citrus trees occurred during fruit set, while it was the least during the winter (Dasberg, 1987). Average concentrations of total N in fruit in our study were 16.3 g N kg^{-1} on April 19, 10.7 g N kg^{-1} on June 8, 8.8 g N kg^{-1} on September 9, and 8.4 g N kg^{-1} on December 1 (Fig. 5.4B). Legaz and Primo-Millo (1988) showed that fruit was the only organ of 4-yr-old 'Valencia' orange trees that consistently accumulated N from the fertilizer or from the N stored in reserve organs, indicating a preferential input; even though, the total N concentration of fruit changed as follows: 22.5 g N kg^{-1} during fruit set, 17.2 during second flush of leaves, and 13.6 at dormancy. Paramasivam et al. (2000) observed that N concentrations of 'Hamlin' orange fruit decreased with their enlargement during June through November. The ^{15}N recovery in the fruit was 10.2 to 18.5% (Table 5.6) which represents the N absorbed from the labeled fertilizer applied in February. Apparent low recovery of added N by fruit is also reported for other fruit crops. Field measurements on mature almond trees showed that 10 to 25% of ^{15}N was recovered in the harvested fruit (Weinbaum et al., 1984). The removal of ^{15}N in harvested kiwi fruit in the first year after N fertilization was about 5 to 6% of the applied labeled fertilizer, even though total removal by the vine tree was about 50% (Ledgard and Smith, 1992). Weinbaum and Kessel (1998) found that total almond tree recovery of applied ^{15}N-depleted N fertilizer during a six-year experimental period was 29.4%. Fruits were the dominant sink, and

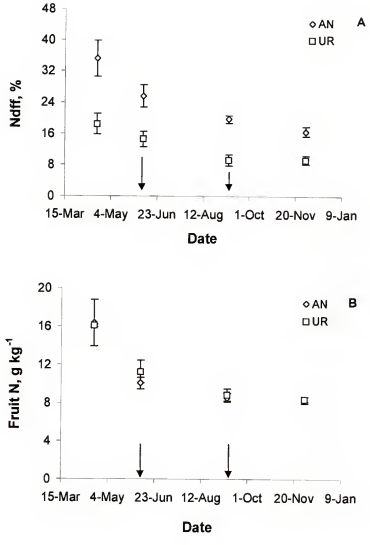

Figure 5.4. Percentage of N derived from the labeled fertilizer (%Ndff) in 'Hamlin' orange fruit applied in the spring (Feb. 15) and total N concentration of fruit over the course of the experiment. Legend: AN = ammonium nitrate treated trees, and UR = urea treated trees. Vertical bars are the standard error of the mean ($n = 3$). Arrows indicate further application of nonlabeled fertilizer. a) Nitrogen in the fruit derived from the labeled fertilizer vs. time; b) N concentration in the fruit vs. time.

Table 5.4. Mean nitrogen concentration of 6-yr-old 'Hamlin' orange trees on Swingle citrumelo rootstock dry matter biomass components, 280 days after ^{15}N labeled ammonium nitrate (AN) or urea (UR) fertilizers application.

Tree component	AN		UR		P < F
	N	sd †	N	sd †	
	g kg^{-1}				
Summer + fall 1999 leaf flush	25.5	1.9	25.5	0.4	0.9779
Spring 1999 + older leaf flush	23.4	1.7	21.2	0.3	0.1001
Twigs >1.5 cm diameter	4.1	0.3	4.0	0.2	0.6873
Twigs ≤1.5 cm diameter	7.8	1.5	7.0	0.7	0.5655
Trunk	4.4	0.2	4.0	0.1	0.0362
Fruit	8.3	0.2	8.4	0.8	0.6898
0-15 cm soil depth					
Fibrous roots (≤0.2 cm diameter)	17.0	2.8	15.2	1.0	0.3633
Woody roots (0.2-1.0 cm diameter)	7.6	1.0	7.1	0.4	0.4600
Woody roots (≥1.0 cm diameter)	6.0	0.5	5.9	1.0	0.8056
15-30 cm soil depth					
Fibrous roots (≤0.2 cm diameter)	15.6	1.3	15.0	1.0	0.6088
Woody roots (0.2-1.0 cm diameter)	8.0	0.4	7.5	1.3	0.5238
Woody roots (≥1.0 cm diameter)	6.4	0.7	7.2	1.6	0.5031
30-45 cm soil depth					
Fibrous roots (≤0.2 cm diameter)	14.0	0.3	15.0	0.1	0.0021
Woody roots (0.2-1.0 cm diameter)	7.6	1.0	7.6	0.9	0.9999
Woody roots (≥1.0 cm diameter)	5.8	0.3	5.8	0.5	0.9999
Taproot	3.8	0.3	3.7	0.1	0.7488

† The standard error of the mean (n = 3).

Table 5.5. Mean nitrogen ^{15}N enrichment of 6-yr-old 'Hamlin' orange trees on Swingle citrumelo rootstock dry matter biomass components, 280 days after ^{15}N labeled ammonium nitrate (AN) or urea (UR) fertilizers application.

Tree component	AN		UR	
	Mean	sd †	Mean	sd †
	Atom% ^{15}N		atom% ^{15}N	
Summer 1999 + fall leaf flush	1.1163	0.2671	0.7876	0.0717
Spring 1999 + older leaf flush	1.2278	0.0993	0.9041	0.1833
Twigs >1.5 cm diameter	0.9745	0.1132	0.7253	0.0660
Twigs ≤1.5 cm diameter	1.0825	0.1436	0.8249	0.0751
Trunk	0.8408	0.1638	0.6231	0.0924
Fruit (9-month-old)	1.9707	0.1171	1.2623	0.1059
0-15cm soil depth				
Fibrous roots (≤0.2 cm diameter)	1.0300	0.0712	0.8269	0.0246
Woody roots (0.2-1.0 cm diameter)	0.8143	0.0120	0.8283	0.0194
Woody roots (≥1.0 cm diameter)	0.8590	0.0022	0.7677	0.1074
15-30cm soil depth				
Fibrous roots (≤0.2 cm diameter)	0.9749	0.1140	0.7511	0.0589
Woody roots (0.2-1.0 cm diameter)	0.9704	0.1404	0.7027	0.1374
Woody roots (≥1.0 cm diameter)	0.9260	0.1116	0.8509	0.0319
30-45cm soil depth				
Fibrous roots (≤0.2 cm diameter)	0.9624	0.0296	0.6454	0.0563
Woody roots (0.2-1.0 cm diameter)	0.8802	0.1252	0.8769	0.1222
Woody roots (≥1.0 cm diameter)	0.8767	0.0995	0.8716	0.1943
Taproot	0.6759	0.1032	0.7289	0.0581

† The standard error of the mean (n = 3).

Table 5.6. Average ^{15}N recovery of 6-yr-old 'Hamlin' orange tree biomass components as calculated by isotopic dilution, 280 days after ^{15}N labeled ammonium nitrate (AN) or urea (UR) fertilizers application.

Tree component	AN		UR		P < F
	N recov.	sd †	N recov.	sd †	
	---------- ^{15}N, % of applied ----------				
Summer + fall 1999 leaf flush	5.49	0.64	3.22	0.20	0.0574
Spring 1999 + older leaf flush	3.08	0.23	1.85	0.21	0.0267
Twigs >1.5 cm diameter	1.09	0.15	0.62	0.09	0.2431
Twigs ≤1.5 cm diameter	4.12	0.84	2.62	0.56	0.0641
Trunk	0.60	0.13	0.37	0.08	0.1814
Fruit (9-month-old)	18.45	1.66	10.24	2.01	0.0148
0-15 cm soil depth					
Fibrous roots (≤0.2 cm diameter)	3.67	0.56	2.25	0.13	0.0587
Woody roots (0.2-1.0 cm diameter)	0.35	0.04	0.33	0.03	0.6681
Woody roots (≥1.0 cm diameter)	0.26	0.20	0.23	0.20	0.8504
15-30 cm soil depth					
Fibrous roots (≤0.2 cm diameter)	1.04	0.39	0.62	0.20	0.1766
Woody roots (0.2-1.0 cm diameter)	0.38	0.21	0.22	0.16	0.3540
Woody roots (≥1.0 cm diameter)	0.16	0.07	0.17	0.10	0.9479
30-45 cm soil depth					
Fibrous roots (≤0.2 cm diameter)	0.13	0.08	0.07	0.04	0.2920
Woody roots (0.2-1.0 cm diameter)	0.03	0.02	0.03	0.01	0.9848
Woody roots (≥1.0 cm diameter)	0.03	0.02	0.03	0.03	0.8244
Taproot	0.58	0.08	0.65	0.08	0.7178
Total	39.45		25.53		0.0062

† The standard error of the mean ($n = 3$).

accounted for 78% of the labeled fertilizer-N recovered by the trees over the period of study.

The fate of added ^{15}N during the spring in different tree components is shown in Fig. 5.5. The largest amount was found in fruit (5.8 and 10.5 g ^{15}N per tree), followed by roots and leaves. The amounts found in woody tissues (i.e., trunk, twigs >1.5 cm diameter, and woody roots) were very low (<0.6 g ^{15}N per tree). Data presented are in agreement with those of Feigenbaum et al. (1987), who reported that the highest percentage of fertilizer-^{15}N was found in the new organs (fruit, twigs and leaves) formed during the previous season for a 22-yr-old Shamouti orange tree. Total roots and trunk plus main branches took up only 2.5 to 4.0% of applied ^{15}N. Thereafter, we verify that the annual growth and the fruit yield of citrus trees contain a small proportion of the fertilizer N applied during the growth period, and the largest portion of N in the new growth is drawn from the tree biomass.

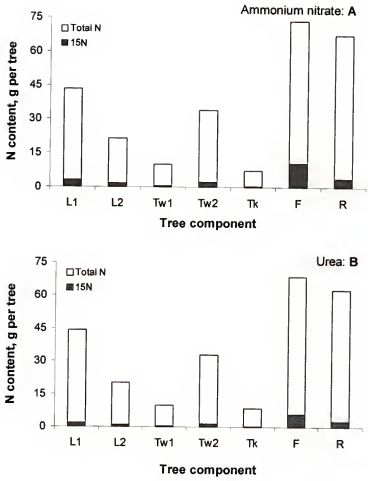

Figure 5.5. Total N and ^{15}N contents of tree components 280 days after application of labeled N as ammonium nitrate or urea. Legend: L1 = summer plus fall 1999 leaf flush; L2 = spring 1999 plus older leaf flush; Tw1= twigs >1.5 cm; Tw2 = twigs ≤1.5 cm; Tk = trunk; F = fruit; and R = total roots within 45 cm soil depth layer. a) Ammonium nitrate treated trees; b) Urea treated trees.

Conclusion

Aboveground biomass of 'Hamlin' orange trees grown on sandy soil with continuous monitored low volume irrigation, expressed as a percentage of the total biomass, was >70%. Fruits were the major component of tree biomass (about 30%). Fibrous roots were concentrated in the 0 to 15 cm soil depth layer, and represented >70% of the total of the below ground class within the 0 to 45 cm layer. Recoveries of ^{15}N by citrus trees fertilized during the spring with ammonium nitrate and urea were 39.5 and 25.5%, respectively. Total ^{15}N recovery was affected by ammonia volatilization, since fertilizers were applied to the soil surface with an alkaline reaction (pH>7). Fruit appeared to be a strong sink for applied ^{15}N (recovery of 10-18%) or redistributed N in the citrus tree. The average ^{15}N content of the tree biomass was small (8% of total N) compared with the total N. The maximum observed quantities were associated with the fruit. About 10.5 and 5.8 g ^{15}N per tree were observed for ammonium nitrate and urea treated trees, respectively.

CHAPTER 6
SUMMARY OF CONCLUSIONS

Field and laboratory evaluations were conducted in two major citrus producing regions in the world to develop a basis for nutrient management of citrus trees with respect to N, P, and K fertilization. The overall conclusions were:

1. Current guidelines for NPK fertilization of nonbearing sweet orange trees (< 5-yr-old) in Brazil should be reviewed, since a differential response of fruit yield to fertilization was observed at higher rates and varied with rootstock cultivars. That is especially important since there has been increased use of other rootstocks besides the Rangpur lime in the Brazilian citriculture.
2. Fertilization (NPK) had only minor effects on canopy volume for trees on the same rootstock after 5 years of tree planting in the field. This suggests that canopy leaf density may more likely be affected by fertilizer rates than canopy volume. Canopy volume was significantly different for the trees on different rootstocks. The trees on Swingle citrumelo rootstock presented the smallest canopy volume as compared to those on Rangpur lime and Cleopatra mandarin rootstocks.
3. Fruit yield of trees on Cleopatra mandarin rootstock increased with P rates (total of 2200 g P2O5 per plant), while that of trees on Swingle citrumelo rootstock increased with K fertilization up to 1800 g K2O per tree applied during a 5-yr-period of fertilization. Maximum fruit yield of trees on Rangpur

lime rootstock was attained with lower NPK rates in comparison to the other rootstocks, except for soils with very low soil exchangeable K level. When soil K levels were very low, a near-linear response of fruit yield to fertilization was observed with K rates up to 1800 g K2O per tree applied during the 5 years of fertilization.

4. Fruit yield was well correlated with leaf N, P, and K concentrations as well as soil P (resin extracted) and exchangeable K. The critical levels of soil and leaf nutrient concentrations for nonbearing trees appeared to be higher than those currently adopted for bearing trees in Brazil.
5. Significant N losses by ammonia (NH3) volatilization were observed from dry granular N fertilizers applied to the soil surface with no incorporation either in soils from Brazil (pH = 5.4) or Florida (pH = 7.0). Gaseous losses of NH3 from urea and ammonium nitrate accounted for up to 44% and 13% of applied N, respectively. Maximum rates of NH3 volatilization occurred within 5 days after fertilizer application.
6. Fertilization with N decreased N mineralization and increased the amount of soil microbial biomass N (MBN) at the 0-15 cm soil depth within 30 days after fertilizer application to the surface of a sandy soil under 6-yr-old citrus trees in Florida. Future studies concerning the fate of N in the soil-plant system should take in account these possible effects.
7. Leaching of N from the 15-30 cm depth layer of soil as estimated by mass balance using the buried PVC column technique was < 9 mg N kg-1 during a 90-day evaluation period. This amount represented about 13% of applied N

fertilizer and was observed when using a low volume irrigation under the tree canopy.

8. The majority of dry matter biomass of 6-yr-old 'Hamlin' orange trees on Swingle citrumelo grown in a sandy Entisol was aboveground (70% of the total biomass). Root dry weight and root length density were concentrated in the surface soil (0-15 cm depth layer). Best management practices should take into account the effective root depth to achieve maximum tree nitrogen uptake and minimize losses in the environment.
9. The total recoveries of 15N by 6-yr-old 'Hamlin' orange trees were 25.5% for urea and 39.5% for ammonium nitrate, at fruit harvest, 280 days after fertilization. The difference of fertilizer N efficiency was attributed to significant gaseous losses of ammonia from urea. Fruit represented a strong sink for applied 15N, which confirms the importance of the spring application of N during early development of fruit. However, the major source of fruit N still appeared to be redistributed N in the citrus tree. Recent leaf flush accumulated more of the 15N applied compared to the older leaves. The accumulation of the labeled N occurred to a lesser extent in woody tissue (twigs, trunk, taproot, and roots > 1.0 cm in diameter), which represented a more steady pool of plant N.

APPENDIX A
SUPPLEMENTAL DATA (Chapter 2)

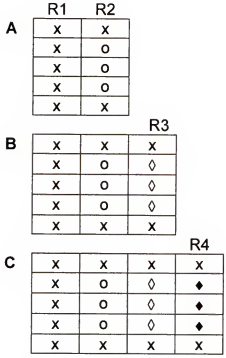

Figure A1. Diagram of experimental plots. Legend: R = planting row; x = buffer tree (Rangpur lime); o = Rangpur lime; ◊ = Cleopatra mandarin; and ♦ = Swingle citrumelo rootstocks. a) Cruz R. Pardo; b) Bebedouro; and c) Matão.

Table A1. Response functions for selected soil chemical characteristics (0-20 cm depth) in the fifth year of the NPK fertilization program.

Soil	Model Coefficients ‡										R^2
parameter †	B_0	N	N^2	P	P^2	K	K^2	NP	NK	PK	
					Sta. Cruz R. Pardo						
pH	Y = 5.1	-0.00113	4.34E-07	0.00067	-6.90E-08	0.00048	-1.50E-07	-1.81E-07	2.27E-07	-2.60E-07	0.40
V, %	Y = 57.0	-0.05486	1.90E-05	0.01799	-9.90E-07	0.02130	-4.02E-06	-4.80E-07	5.79E-06	-1.22E-05	0.45
P-res, mg kg^{-1}	Y = 71.7	-0.08806	3.42E-05	0.03230	1.70E-07	0.03430	-5.25E-06	1.26E-05	-2.31E-05	-1.03E-05	0.40
K, mmol dm^{-3}	Y = 0.7	-0.00002	8.70E-09	-0.00068	2.69E-07	0.00103	-1.38E-07	-8.47E-08	-2.83E-08	2.25E-07	0.73
					Bebedouro						
pH	Y = 4.8	-0.00077	2.26E-07	0.00020	1.39E-07	0.00026	5.00E-08	-1.94E-07	7.30E-08	-2.77E-07	0.78
V, %	Y = 32.0	-0.02004	6.34E-06	0.02069	-2.60E-07	0.01513	-1.63E-06	-7.10E-06	2.32E-06	-8.58E-06	0.75
P-res, mg kg^{-1}	Y = 12.1	-0.04902	1.33E-05	0.05562	-1.48E-05	0.02168	-2.24E-05	6.96E-06	9.88E-06	1.12E-05	0.58
K, mmol dm^{-3}	Y = 1.1	-0.00105	1.48E-07	0.00079	-1.48E-07	0.00257	-3.37E-07	2.35E-07	-4.50E-08	-5.02E-07	0.45

Table A1. Continued

Soil parameter †	Model Coefficients ‡										R^2
	B_0	N	N^2	P	P^2	K	K^2	NP	NK	PK	
					Matão						
pH	Y = 5.0	-0.00057	1.74E-07	0.00007	-1.70E-08	-0.00006	7.50E-08	-3.30E-08	-8.30E-08	-1.00E-08	0.61
V, %	Y = 56.5	-0.08269	2.53E-05	0.03870	-1.12E-05	0.00303	-3.80E-07	1.78E-05	-5.28E-06	6.20E-07	0.60
P-res, mg kg^{-1}	Y = 0.5	0.00034	6.94E-08	-0.00015	2.08E-07	0.00302	-1.50E-07	-2.75E-07	-6.83E-07	8.00E-08	0.79
K, mmol dm^{-3}	Y = 55.5	-0.02265	8.16E-06	-0.00368	3.13E-06	0.01010	2.50E-07	-5.80E-07	-5.13E-06	-4.50E-06	0.60

† P-res: anionic exchangeable resin; K: exchangeable potassium; pH: soil / $CaCl_2$ 0.01 mol L^{-1}, 1:2.5 ratio (v/v); V: base saturation.
‡ Rates of NPK are in g N $tree^{-1}$, g P_2O_5 $tree^{-1}$, or g K_2O $tree^{-1}$.

APPENDIX B
SUPPLEMENTAL DATA (Chapter 5)

Table B1. Water content of biomass components of 6-yr-old 'Hamlin' orange trees on Swingle citrumelo rootstock treated with ammonium nitrate.

Tree component	Water content	sd †	cv ‡
	---------- g kg^{-1} dry matter ----------		
Summer + fall 1999 leaf flush	1686	87	51
Spring 1999 + older leaf flush	1491	106	71
Twigs >1.5 cm diameter	683	06	08
Twigs ≤1.5 cm diameter	812	44	55
Trunk	676	66	97
Fruit (9-month-old)	5913	388	66
SE Quadrant: 0-15 cm soil depth			
Fibrous roots ≤0.2 cm diameter)	2052	16	08
Woody roots (0.2-1.0 cm diameter)	1090	68	62
Woody roots (≥1.0 cm diameter)	865	77	89
SE Quadrant:15-30 cm soil depth			
Fibrous roots (≤0.2 cm diameter)	2050	261	127
Woody roots (0.2-1.0 cm diameter)	1076	85	79
Woody roots (≥1.0 cm diameter)	918	16	18
SE Quadrant: 30-45 cm soil depth			
Fibrous roots (≤0.2 cm diameter)	1524	49	32
Woody roots (0.2-1.0 cm diameter)	1040	56	54
Woody roots (≥1.0 cm diameter)	854	138	162

Table B1. Continued.

Tree component	Water content	sd †	Cv ‡
	---------- g kg^{-1} dry matter ----------		
NW Quadrant: 0-15 cm soil depth			
Fibrous roots (≤0.2 cm diameter)	1949	177	91
Woody roots (0.2-1.0 cm diameter)	1033	51	50
Woody roots (≥1.0 cm diameter)	985	26	27
NW Quadrant: 15-30 cm soil depth			
Fibrous roots (≤0.2 cm diameter)	2096	238	114
Woody roots (0.2-1.0 cm diameter)	1025	34	34
Woody roots (≥1.0 cm diameter)	851	171	201
NW Quadrant: 30-45 cm soil depth			
Fibrous roots (≤0.2 cm diameter)	2401	311	130
Woody roots (0.2-1.0 cm diameter)	1074	132	123
Woody roots (≥1.0 cm diameter)	-	-	-
Taproot	556	35	62

† The standard error of the mean ($n = 3$).
‡ Coefficient of variation.

Table B2. Water content of biomass components of 6-yr-old 'Hamlin' orange trees on Swingle citrumelo rootstock treated with urea.

Tree component	Water content	sd †	cv ‡
	---------- g kg^{-1} dry matter ----------		
Summer + fall 1999 leaf flush	1647	35	22
Spring 1999 + older leaf flush	1442	12	08
Twigs >1.5 cm diameter	560	50	89
Twigs ≤1.5 cm diameter	651	75	115
Trunk	577	66	114
Fruit (9-month-old)	5941	153	26

† The standard error of the mean (n = 3).
‡ Coefficient of variation.

Table B3. Total N content of biomass components of 6-yr-old 'Hamlin' orange trees treated with ammonium nitrate (AN) or urea (UR) as determined for samples collected in December 1999.

Tree component	AN		UR	
	N	sd †	N	sd †
	g			
Summer 1999 leaf flush	40.16	8.53	42.36	3.36
Spring 1999 + older leaf flush	19.72	1.64	19.02	1.04
Twigs > 15 mm diameter	9.49	4.32	9.58	0.57
Twigs ≤15 mm diameter	31.59	3.82	31.39	1.46
Trunk	6.90	0.67	8.18	2.06
Fruit (9-month-old)	63.16	2.05	62.72	15.61
0-15 cm soil depth				
Fibrous roots (≤0.2 cm diameter)	30.07	4.65	26.94	1.01
Woody roots (0.2-1.0 cm diameter)	4.28	0.54	4.00	0.54
Woody roots (≥1.0 cm diameter)	2.89	2.21	2.89	2.36
15-30 cm soil depth				
Fibrous roots (≤0.2 cm diameter)	9.31	2.73	9.06	2.88
Woody roots (0.2-1.0 cm diameter)	3.33	1.58	3.19	1.73
Woody roots (≥1.0 cm diameter)	1.70	0.81	1.92	1.09
30-45 cm soil depth				
Fibrous roots (≤0.2 cm diameter)	1.22	0.78	1.31	0.83
Woody roots (0.2-1.0 cm diameter)	0.36	0.17	0.36	0.18
Woody roots (≥1.0 cm diameter)	0.31	0.28	0.33	0.31
Tap root	10.17	3.81	9.87	3.08
Total	234.67		233.12	

† The standard error of the mean (n=3).

Table B4. Total ^{15}N content of biomass components of 6-yr-old 'Hamlin' orange trees treated with labeled ammonium nitrate (AN) or urea (UR) as determined for samples collected in December 1999

Tree component	AN		UR	
	^{15}N	sd †	^{15}N	sd †
	g			
Summer 1999 leaf flush	3.13	0.99	1.83	0.11
Spring 1999 + older leaf flush	1.76	0.13	1.05	0.33
Twigs > 15 mm diameter	0.62	0.34	0.35	0.05
Twigs ≤15 mm diameter	2.35	0.60	1.50	0.32
Trunk	0.34	0.11	0.21	0.09
Fruit (9-month-old)	10.51	0.95	5.84	1.73
0-15 cm soil depth				
Fibrous roots (≤0.2 cm diameter)	2.09	0.53	1.28	0.07
Woody roots (0.2-1.0 cm diameter)	0.20	0.02	0.19	0.02
Woody roots (≥1.0 cm diameter)	0.15	0.11	0.13	0.12
15-30 cm soil depth				
Fibrous roots (≤0.2 cm diameter)	0.59	0.22	0.36	0.11
Woody roots (0.2-1.0 cm diameter)	0.22	0.12	0.13	0.09
Woody roots (≥1.0 cm diameter)	0.09	0.04	0.10	0.06
30-45 cm soil depth				
Fibrous roots (≤0.2 cm diameter)	0.07	0.05	0.04	0.02
Woody roots (0.2-1.0 cm diameter)	0.02	0.01	0.02	0.01
Woody roots (≥1.0 cm diameter)	0.02	0.01	0.02	0.02
Tap root	0.33	0.14	0.37	0.12
Total	22.49		13.41	

† The standard error of the mean (n=3).

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BIOGRAPHICAL SKETCH

Born in Neves Paulista, Brazil, in 1966, Dirceu started undergraduate course in agronomic engineering at the São Paulo State University (UNESP) in 1985. After completing the B.S. degree in 1990, he became a trainee at the Soil Fertility and Plant Nutrition Section of the Instituto Agronômico (IAC), working in research projects related to soil nitrogen. That same year he was admitted into the master's degree program at the University of São Paulo (USP). Before completing his M.S. program in 1993, he was hired by a private company to direct a soil, plant, and orange juice analysis laboratory. In 1995, he joined the Sylvio Moreira Citrus Research Center of IAC. Since then, he has been engaged in soil fertility and citrus nutrition research. In 1997, he came to the Soil and Water Science Department at the University of Florida to pursue his Ph.D. program and in December 2000 a dream comes true with his degree. Now, he plans to take a month on vacation faraway from laboratories, books and computers to enjoy his family in a wonderful island of the east coast of Brazil.

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

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